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ERRATA

Page 207, legend of figure 2, "ammonia" should read "amino."

Page 221, table 1, lines 3, 6, 9, 12 and 15 of body of table, $Ca(H_2PO_4)_2.2H_2O2H_2O$ should read $Ca(H_2PO_4)_2.2H_2O$.

Page 390, table at top of page, first line of table, "Limed" should read "Not limed," and second line of table, "Not limed" should read "Limed."

SOME AVAILABILITY STUDIES WITH AMMONIUM PHOSPHATE AND ITS CHEMICAL AND BIOLOGICAL EFFECTS UPON THE SOIL[1]

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INTRODUCTION

The purpose in beginning the work which is reported on the following pages was to make a study of the availability and the effect on germination and biological activities of an ammonium phosphate fertilizer manufactured by the American Cyanamid Company. In the preparation of this fertilizer ground phosphate rock is mixed with sufficient sulfuric acid to convert the phosphorus compounds into free phosphoric acid, and the latter removed from the mixture by filtration and washing the residue. Ammonia is produced in gaseous form by steaming crude calcium cyanamid in an autoclave under a pressure of several atmospheres. The ammonia is bubbled into the phosphoric acid until it is converted chiefly into di-ammonium phosphate but at the same time some citrate-insoluble iron and aluminum ammonium phosphates are formed. With methyl orange as an indicator, phosphoric acid is then added until the solution reaches the proportions of mono-ammonium phosphate, the insoluble phosphates being thereby rendered available. The solution after evaporation to dryness is known commercially as "Ammo-Phos." It is a light gray material resembling acid-phosphate in appearance.

A large amount of work has been done with ammonium sulfate, and its relative value as compared with other common nitrogenous fertilizers determined, but for the most part other ammonium salts have received little consideration. Ammonium phosphate, for example, has been omitted from fertilizer studies because it has been considered too expensive to be worthy of consideration as a fertilizer. Now that a satisfactory method has been developed for its manufacture, it is highly important that we know under just what conditions this new fertilizer will give the best results and how much may be safely and economically applied to various crops. Does the ammonia act like that in ammonium sulfate and the phosphorus like that in acid phosphate, as we might expect, or does the combining of the two radicals change their efficiency or chemical effects to any extent? Both the acid and basic

[1] A portion of a thesis submitted to the faculty of Rutgers College in partial fulfillment of the requirements for the degree of Doctor of Philosophy, 1917.

radicals are utilized as foods for plants and for this reason, if no other, we should expect that the effects would be somewhat different, especially after long continued use.

HISTORICAL

Ammonium phosphate was used by several of the early investigators in their studies on the nutrition of plants. These various workers observed that ammonium salts often failed to nourish certain plants as they should, and sought after the reasons for their negative results and the conditions under which ammonium salts could be profitably used. A few investigators have worked on the chemical and biological effects produced by an application of ammonium phosphate to a soil. The effect on germination and young plants has been reported in a few cases. For the most part, though, ammonium phosphate has received little consideration as a possible fertilizer. The work noted by the writer is briefly abstracted below.

Johnson (11) reports the work of Ville who grew wheat in calcined sand, to which was added 0.11 gm. of nitrogen in each case, either in the form of potassium nitrate, ammonium chloride, ammonium nitrate or ammonium phosphate. The results were as follows:

SOURCE OF NITROGEN	STRAW AND ROOTS	GRAIN	AVERAGE CROP	NITROGEN IN CROP
Potassium nitrate	20.70 19.22	6.20 7.30	26.71	0.221
Ammonium chloride	15.10 17.34	4.93 3.54	18.83	0.142
Ammonium nitrate	12.20 14.87	3.72 5.86	18.32	0.133
Ammonium phosphate	12.96 15.82	3.77 4.34	18.40	0.133

It is seen that all the ammonium salts gave about the same results, the crop being about one-fourth less than with potassium nitrate. However, no allowance was made for the potassium in the potassium nitrate.

Hellriegel (11) obtained a larger yield of clover with ammonium sulfate and ammonium phosphate than with ammonium nitrate or sodium nitrate.

Birner and Lucanus (11), in 1864, found that in water cultures sulfate and phosphate of ammonia did not effectively replace nitrates.

Hampe (11), in 1866, using ammonium phosphate and keeping the solution faintly acid, obtained a corn plant with a dry weight of 18 gm. and having 36 perfect seeds. The solution did not contain any nitrates.

Kühn (11), in 1866, grew two small corn plants, one with ammonium phosphate and the other with ammonium sulfate as the only sources of nitrogen. The experiments were interrupted by the high temperature in the greenhouse.

Wagner (11), in 1868, found in agreement with the work of Hampe of the

previous year that a maize seedling, after growing for 2 weeks in an artificial soil grew normally when placed in a nutritive solution containing ammonium phosphate as a source of nitrogen. One plant, hen dried, weighed 26.5 gm. and carried 48 ripe seeds.

Claudel and J. Cochetelle (3) found that basic phosphates, including ammonium phosphate, are favorable to germination and especially to leguminous seeds. Potassium salts, ammonium sulfate, sodium nitrate and superphosphate were injurious to germination, especially to lentils, lucerne and flax. Wheat was quite resistant.

Tanret (33), using nutrient solutions containing an excess of either ammonium nitrate, ammonium sulfate or ammonium chloride for the culture of *Aspergillus niger* found that no spores were produced during the first month of growth. On the other hand, ammonium phosphate favored spore production.

Steffeck and Maercker (32), using ammonium phosphate as a nitrogenous manure, found that it acted in a manner similar to ammonium sulfate with both oats and barley, both as regards the yield and nitrogen content of the grain. As a phosphate fertilizer the ammonium phosphate yielded only 88.33 per cent as much as acid phosphate soluble in water.

Hall and Gimingham (6) shook soil for 24 hours with a solution of ammonium salts and determined whether there was an increase in acidity or evolution of ammonia. Sand gave negative results. Clay showed no change in acidity or withdrawal of the acid radical from solution. Complete absorption of the base did not occur in clay with ammonium chloride, ammonium carbonate or ammonium phosphate. In the case of the latter two salts some of the acid was removed from solution.

Pantanelli and Severini (27), growing wheat, mustard, corn, flax and rice in water cultures, found that a rapid absorption of the ammonium cation increases the acidity of the nutritive medium which reaches a maximum during the first period of growth for those salts of ammonium derived from strong acids, including phosphoric acid. After the first week of growth the acidity decreases if the anion is absorbed rapidly, as is the case with nitric and phosphoric acids. If the ammonium salt is only slightly soluble, as is the case with ammonium magnesium phosphate, the acid production is avoided.

Rusche (29) concluded from his work with various crops, using 10 gm. of salt per 11.5 kgm. of soil, that ammonium phosphate usually has a good effect, working injuriously only with clover, serradella and rape. All phosphate salts showed some injury to the germination of seeds. With beets the greatest root system was produced by sulfates and the smallest by ammonium nitrate, ammonium phosphate, calcium nitrate, and sodium phosphate. Ammonium phosphates proved injurious to the root systems of alfalfa. The ammonium salts in general were quite injurious. The influence of ammonium phosphate on the parts of the plants above ground was good. Small weights were obtained with chlorides and sulfates with the exception of ammonium sulfate which, like the ammonium phosphate, had a good effect.

EXPERIMENTAL

The experimental work includes the analysis of "Ammo-Phos," biological studies, and various availability experiments in the field and greenhouse, including the effect on lime-requirement and germination. The methods used

TABLE 1

Composition of "Ammo-Phos"

	PER CENT
Moisture	4.55
SiO_2	0.50
Al_2O_3	2.30
Fe_2O_3	4.50
K_2O	0.33
Na_2O	1.35
Cl	0 10
CaO	2.52
MgO	0.31
NH_3	13.54
Nitrate	Trace
Total P_2O	43.13
Water-soluble P_2O_5	36.80
Citrate-soluble P_2O_5	4.82
Citrate-insoluble P_2O_5	1.51

TABLE 2

Composition of "Ammo-Phos"

	COMPLETE "AMMO-PHOS"	WATER-SOLUBLE PORTION OF "AMMO-PHOS"	WATER-INSOLUBLE PORTION OF "AMMO-PHOS"	CITRATE-INSOLUBLE PORTION OF "AMMO-PHOS"
Moisture at 100°C	7.14	0.00	1.04	0.84
NH_3	13.58	16.43	6.36	5.51
Water-soluble P_2O_5	35.16	48.98	0.00	0.00
Citrate-soluble P_2O_5	5.76	0.00	37.02	0.00
Citrate-insoluble P_2O_5	1.10	0.00	2.84	44.19
Total P_2O_5	42.02	48.98	39.86	44.19
SO_3	Not det.	Not det.	Trace	0 00
Ca	Not det.	Not det.	1.20	Trace
Fe_2O_3	Not det.	Trace	19.00	Not det.
Al_2O_3	Not det.	Not det.	8.50	Not det.
$Fe_2O_3 + Al_2O_3$	Not det.	Not det.	27.50	23.00
Loss on ignition (red heat)	Not det.	Not det.	18.14	Not det.

varied for the different experiments and are discussed as the data are given throughout the thesis.

ANALYTICAL

A representative sample of the commercial ammonium phosphate ("Ammo-Phos"), made from Tennessee rock phosphate was ground and analyzed for

the more common elements that it seemed probable would be found in the product. The composition in per cent is given in table 1.

An analysis made by the American Cyanamid Company of similar material is given in table 2.

These two analyses bring out the high plant-food content and in addition show that practically all of the phosphorus and nitrogen are present in a readily available form. In addition to containing slightly more than two-thirds as much nitrogen as does sodium nitrate, there is also present in any given quantity nearly three times as much phosphoric acid as is present in a like weight of acid phosphate. As far as the analyses would indicate there are no constituents present that are likely to prove injurious to plant growth.

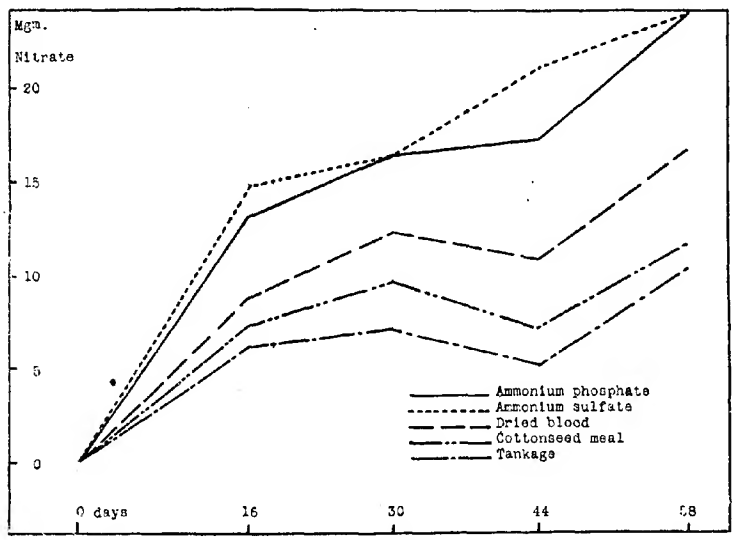

FIG. 1. DIAGRAM SHOWING THE NITRIFICATION OF VARIOUS FERTILIZERS IN FRESH SOIL

BIOLOGICAL

The purpose of the biological studies was primarily to determine the relative availability of ammonium phosphate under different soil conditions as compared with other nitrogenous fertilizers. It is a well known fact that organic fertilizers are rapidly nitrified when placed in the soil under favorable conditions for the action of bacteria and fungi but the formation of nitrates is not as rapid as with ammonium sulfate. Will ammonium phosphate be as rapidly transformed as ammonium sulfate? We should expect this to be the case but possibly the difference in the acid radical would play an important rôle in various soils.

TABLE 3

Nitrification of various fertilizers in fresh soil

TREATMENT	16 DAYS				30 DAYS				44 DAYS				58 DAYS			
	Nitrate nitrogen	Average	Increase over check	Per cent nitrified	Nitrate nitrogen	Average	Increase over check	Per cent nitrified	Nitrate nitrogen	Average	Increase over check	Per cent nitrified	Nitrate nitrogen	Average	Increase over check	Per cent nitrified
	mgm.	*mgm.*	*mgm.*		*mgm.*	*mgm.*	*mgm.*		*mgm.*	*mgm.*	*mgm.*		*mgm.*	*mgm.*	*mgm.*	
Check	1.95 1.95	1.95			1.25 1.04	1.15			0.79 0.74	0.76			2.33 2.47	2.40		
Ammonium phosphate	14.28 16.17	15.22	13.27	62.60	18.37 17.48	17.93	16.78	79.20	17.37 18.68	18.02	17.26	81.80	26.77 26.41	26.59	24.19	114.81
Ammonium sulfate	18.00 15.88	16.94	14.99	71.00	17.99 17.73	17.86	16.71	78.80	20.28 24.06	22.18	21.42	101.50	25.00 24.69	24.84	22.44	106.30
Dried blood	10.90 11.25	11.07	9.12	43.00	14.29 13.01	13.65	12.50	59.00	11.75 12.25	12.00	11.24	53.20	18.89 20.16	19.52	17.12	81.20
Cottonseed meal	9.75 9.25	9.50	7.55	35.60	11.09 11.23	11.16	10.01	47.20	7.89 8.98	8.43	7.67	36.40	15.19 13.89	14.54	12.14	57.50
Tankage	8.13 8.75	8.44	6.49	30.60	8.42 8.93	86.80	7.53	35.50	6.58 6.31	6.44	5.68	26.90	13.12 13.53	13.32	10.92	51.30

Comparative availability of ammonium phosphate with other nitrogenous fertilizers as shown by nitrification

The method adopted was the ordinary tumbler-fresh-soil method, 100-gm. portions of a loam soil being used. In addition to ammonium phosphate, ammonium sulfate, dried blood, cottonseed meal and tankage were used, each tumbler receiving the same amount of nitrogen as that present in 100 mgm. of ammonium sulfate. The soil used was slightly acid and in order to make conditions more favorable for the nitrifying bacteria 0.5 gm. of ground limestone was added to each tumbler. The fertilizers were thoroughly mixed with the soil by means of a soil shaker (16). The tumblers were then allowed to incubate at room temperature for varying lengths of time, as shown in table 3, and the nitrates extracted by leaching from the soil with 500 cc. of water and determined in an aliquot of the filtrate by the phenol-disulfonic acid method. The data are given in table 3 and shown graphically in figure 1.

It will be noticed that ammonium phosphate compares very favorably with ammonium sulfate and the other nitrogenous fertilizers used. The data show that in this particular soil ammonium phosphate does not nitrify quite as rapidly as ammonium sulfate but after one month there is little difference. When compared with dried blood, cottonseed meal and tankage, ammonium phosphate shows a much higher availability. The relative positions of these last-named materials, when compared with the ammonium phosphate fertilizer or with each other, does not change, regardless of the incubation period.

In three cases nitrification of the fertilizer added amounted to more than 100 per cent, doubtless because of the inaccuracy of the phenol-disulfonic acid method in the presence of soluble organic matter, various soluble salts and large amounts of nitrate nitrogen. It is barely possible that the fertilizer stimulated the nitrification of the organic matter of the soil, but this is improbable.

Comparative rate of nitrification of nitrogenous fertilizers in different soils

This experiment was carried out in tumblers with fresh soil according to the method previously given, equivalent amounts of nitrogen (20.7 mgm. per tumbler) being applied in each case. The two soils used were a Penn loam taken from a very fertile garden and a Sassafras loam of medium fertility taken from a meadow. Both soils were slightly acid but 0.5 per cent of calcium carbonate was added to all tumblers to neutralize the acidity. The results obtained are given in table 4 and shown diagrammatically in figures 2 and 3.

It will be noticed that in the garden soil, as in the case of experiments previously reported, ammonium phosphate and ammonium sulfate are both rapidly nitrified and at about the same rate. The differences at the various incubation periods are so slight that they may almost be considered within

TABLE 4

Rate of nitrification of fertilizers in soils

TREATMENT	GARDEN LOAM				MEADOW LOAM			
	Nitrate nitrogen	Average	Increase over check	Per cent nitrified	Nitrate nitrogen	Average	Increase over check	Per cent nitrified
2 weeks								
	mgm.	*mgm.*	*mgm.*		*mgm.*	*mgm.*	*mgm.*	
Acid phosphate	0.72 0.69	0.71			0.52 0.44	0.48		
Ammonium phosphate	9.03 8.89	8.96	8.25	39.85	5.56 6.67	6.11	5.63	27.19
Ammonium sulfate and acid phosphate	6.94 8.35	7.63	6.92	33.43	4.17 4.17	4.17	3.69	17.77
Dried blood and acid phosphate	4.30 4.72	4.51	3.80	18.36	4.03 3.61	3.82	3.34	16.13
Cottonseed meal and acid phosphate	4.72 4.86	4.79	4.08	19.71	2.91 2.99	2.95	2.47	11.93
4 weeks								
Acid phosphate	1.02 0.97	1.00			0.66 0.63	0.65		
Ammonium phosphate	15.29 18.07	16.68	15.68	75.75	11.95 11.12	11.53	10.88	52.56
Ammonium sulfate and acid phosphate	16.68 18.07	17.37	16.37	79.08	13.07 11.12	12.09	11.44	55.26
Dried blood and acid phosphate	10.84 9.73	10.28	9.28	44.83	11.40 9.73	10.56	9.91	47.87
Cottonseed meal and acid phosphate	lost 9.73	9.73	8.73	42.17	6.12 6.67	6.39	5.74	27.72
6 weeks								
Acid phosphate	1.63 1.65	1.64			0.82 0.75	0.79		
Ammonium phosphate	22.80 23.07	22.93	21.29	102.85	12.23 13.90	13.06	12.27	59.27
Ammonium sulfate and acid phosphate	23.07 21.96	22.51	20.87	100.82	14.73 13.90	14.31	13.52	65.31
Dried blood and acid phosphate	16.40 13.90	15.15	13.51	65.26	11.12 lost	11.12	10.33	49.90
Cottonseed meal and acid phosphate	12.79 12.79	12.79	11.15	53.86	7.23 8.06	7.64	6.85	33.09

TABLE 4—(Concluded)

TREATMENT	GARDEN LOAM				MEADOW LOAM			
	Nitrate nitrogen	Average	Increase over check	Per cent nitrified	Nitrate nitrogen	Average	Increase over check	Per cent nitrified
8 weeks								
	mgm.	*mgm.*	*mgm.*		*mgm.*	*mgm.*	*mgm.*	
Acid phosphate	1.32 1.42	1.37			0.86 0.88	0.87		
Ammonium phosphate	19.11 19.33	19.44	18.07	83.32	13.77 13.77	13.77	12.90	62.31
Ammonium sulfate and acid phosphate	19.11 18.89	19.00	17.63	81.24	14.44 15.11	14.77	13.90	67.15
Dried blood and acid phosphate	11.33 12.00	11.66	10.29	45.57	12.22 11.55	11.88	11.01	53.18
Cottonseed meal and acid phosphate	8.99 8.66	8.82	7.45	34.33	7.33 6.66	6.99	6.12	29.56
10 weeks								
Acid phosphate	1.73 1.73	1.73			0.69 0.78	0.73		
Ammonium phosphate	16.44 17.77	17.10	15.37	74.25	15.11 15.33	15.22	14.49	70.00
Ammonium sulfate and acid phosphate	15.31 14.88	15.19	13.46	65.02	14.22 15.33	14.77	14.04	67.82
Dried blood and acid phosphate	10.22 10.44	10.33	8.60	41.55	10.22 11.11	10.66	9.93	47.97
Cottonseed meal and acid phosphate	8.99 8.99	8.99	7.26	35.07	7.77 7.11	7.44	6.71	32.89

experimental error. Dried blood and cottonseed meal run very nearly together and are much more slowly nitrified than the two ammonium fertilizers. Maximum nitrification was obtained in all cases after 6 weeks' incubation. After this, small amounts of nitrate nitrogen were found because the nitrates had been utilized by bacteria and fungi in the soil for their own growth and consequently were changed over to protein. This protein in turn is nitrified after the microörganisms die.

The results obtained with the meadow soil are in general the same, except that nitrification does not proceed at as rapid a rate as in the garden soil, and the total accumulation of nitrates at any one time is never as large. Ammonium phosphate and ammonium sulfate run close together for the most part, but not quite as consistently so as in the richer soil. Cottonseed meal for

some unknown reason ranks low, while dried blood shows a comparatively high availability as measured by nitrification. Maximum nitrification is obtained in the case of cottonseed meal at the end of 6 weeks, dried blood at the end of 8 weeks and the two ammonium fertilizers at the end of 10 weeks.

The work reported in tables 3 and 4, in so far as it is comparable, agrees in a general way with that of Müntz (26) who in comparative tests of the rate of nitrification of various nitrogenous fertilizers found that at the end of 5 months the relative standings were ammonium sulfate 100, calcium cyanamid 88, dried blood 66 and leather 26.

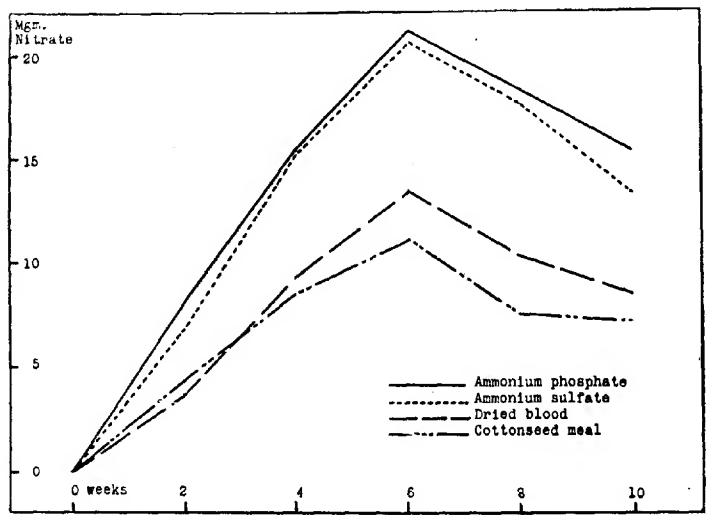

FIG. 2. DIAGRAM SHOWING THE RATE OF NITRIFICATION IN A GARDEN LOAM SOIL

Lipman, Brown and Owen (19) in nitrification experiments, after an incubation period of 4 weeks, obtained the following percentages of recovery of nitrogen as nitrate from various sources of nitrogen:

	per cent
Ammonium sulfate	78.47
Concentrated tankage	53.54
Calcium cyanamid	52.56
Cottonseed meal	30.64
Ground fish	26.15
Dried blood	26.13
Cow manure (solid and liquid, fresh)	12.67
Bone meal	12.31
Cow manure (solid, fresh)	9.46
Horse manure	8.76
Cow manure (solid and liquid, leached)	4.48

The same investigators (20) in another nitrification experiment, after an incubation of 7 weeks, obtained the nitrogen recovery noted below:

	per cent
Ammonium sulfate	73.30
Linseed meal	49.07
Cottonseed meal	42.87
Soybean meal	42.41
Cowpea	33.12
Wheat flour	23.15
Rye flour	13.84

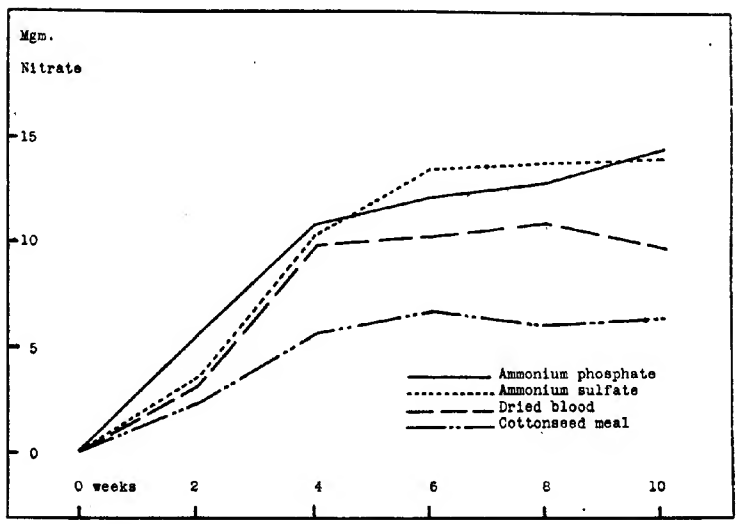

FIG. 3. DIAGRAM SHOWING THE RATE OF NITRIFICATION IN A MEADOW LOAM SOIL

In these experiments the incubation period was for such a long time that no doubt the maximum nitrate production in the case of ammonium sulfate, at least, had been reached prior to the end of the experiment.

The work reported in tables 3 and 4, as well as that of Lipman and that of Müntz, just discussed, brings out very strikingly that as far as the organic forms of nitrogen are concerned the nitrogen-carbon ratio certainly plays an important rôle. The wider the ratio the more difficult it is for the micro-organisms to break down the compounds, and furthermore, the fungi are stimulated to a greater extent. These latter organisms as well as a large portion of the bacteria utilize the nitrates produced for their own growth, and consequently only a comparatively small proportion of the nitrate actually formed from substances of very wide nitrogen-carbon ratio may accumulate. In fact, the added material may actually decrease the amount of nitrates in the soil. On the other hand, ammonium sulfate and ammonium phosphate,

when added to the soil, do not have such a marked effect upon the numbers of organisms, and furthermore, the processes of transformation are less complex. Organic fertilizers must be broken up into peptones, albumoses, amino acids and ammonia, and these in turn nitrified. The process is much longer and requires more energy than the mere converting of ammonia into nitrate.

A discussion of nitrification as a method for the determination of availability would not be complete without referring to the work of Lipman (17) which is in direct contrast with most of the nitrification experiments which have been reported. Using arid soils he found that low-grade nitrogenous materials and ammonium sulfate nitrify much more rapidly than in humid soils, while on the other hand dried blood and other high-grade nitrogenous fertilizers nitrify slowly in these soils. The relative availabilities of the fertilizers were tested out in vegetation experiments and these results confirmed the nitrification data. Considering sodium nitrate as 100, the yields of barley in sand were 82 for calcium nitrate, 139 for ammonium sulfate, 101 for dried blood, 134 for cottonseed meal and 119 for steamed bone meal. Lipman states that "the nitrification method in the direct soil culture, for determining the relative availabilities of nitrogenous fertilizers of the organic or ammonia type, was feasible and more logical and specific in nature than other methods employed heretofore." Under humid conditions other investigators also seem to have obtained reasonably close agreement between nitrification and vegetation tests.

Effect of different forms of lime on the rate of nitrification in soils

The soils selected for this experiment were: first, a very acid sandy loam, so acid in fact that crops did not grow on it except under the most favorable climatic conditions; second, a heavy silt soil only moderately acid and growing good crops each year; and third, a shale loam, fairly fertile and neutral to litmus paper.

The methods used were practically the same as in the nitrification experiments already discussed, 100 gm. of fresh soil per tumbler being used, and the materials thoroughly mixed with the entire sample. Where ammonium sulfate was applied 100 mgm. were added to each tumbler and where ammonium phosphate was used enough was added to supply the same amount of nitrogen as is present in the ammonium sulfate. To all tumblers except the checks and those receiving ammonium phosphate, 586 gm. of acid phosphate were added, thus making the amount of phosphorus the same as in the ammonium phosphate tumblers. Calcium carbonate, where used, was applied at the rate of 1 gm. per tumbler and calcium oxide at the rate of 560 mgm. The incubation period was 26 days. The results are given in table 5 and shown diagrammatically in figure 4.

An examination of this table brings out several points of interest. First, we notice, as has often been shown, that the addition of acid phosphate in-

creases the rate of nitrification of the soil organic matter. The use of calcium oxide, except in the most acid soil, produced a greater stimulation in nitrate production from the soil organic matter than did calcium carbonate. Where ammonium sulfate was applied to the very acid sandy loam soil in the presence of acid phosphate equivalent to that present in ammonium phosphate, the nitrate production was approximately two-thirds of that where ammonium

TABLE 5

Effect of different forms of lime on the rate of nitrification of ammonium phosphate and ammonium sulfate

TREATMENT	SANDY LOAM		SILT		SHALE LOAM	
	Nitrate nitrogen	Average	Nitrate nitrogen	Average	Nitrate nitrogen	Average
	mgm.	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*
Check	0.79 0.79	0.79	0.79 1.05	0.92	0.79 0.92	0.85
Acid phosphate	1.10 1.05	1.07	1.18 1.05	1.11	0.95 0.95	0.95
Calcium oxide and acid phosphate	3.33 3.19	3.26	3.53 3.24	3.38	4.47 5.00	4.73
Calcium carbonate and acid phosphate	3.53 3.82	3.67	1.32 1.39	1.35	1.66 1.93	1.79
Ammonium sulfate and acid phosphate	1.11 1.11	1.11	7.89 7.94	7.91	10.00 9.65	9.82
Ammonium phosphate	1.94 1.77	1.85	7.91 7.68	7.79	11.67 10.83	11.25
Ammonium sulfate, calcium oxide and acid phosphate	3.28 3.19	3.23	1.85 1.39	1.62	13.39 12.99	13.19
Ammonium phosphate and calcium oxide	2.36 2.36	2.36	2.05 2.89	2.47	10.26 11.17	10.71
Ammonium sulfate, calcium carbonate and acid phosphate	13.23 13.53	13.37	10.88 10.58	10.73	16.65 14.43	15.54
Ammonium phosphate and calcium carbonate	13.81 13.41	13.61	11.84 11.56	11.71	17.11 16.81	16.96

phosphate was used alone, but in both cases the amount of nitrate was only slightly above that in the checks. In the silt loam the difference between the two fertilizers in the absence of lime was within experimental error, but the increases over the checks were very great, showing that the soil was not badly in need of lime. In the shale loam there was a slightly greater accumulation of nitrate from ammonium phosphate than from ammonium sulfate, the difference being approximately 14 per cent in favor of the former fertilizer.

As was previously stated, this soil was neutral to litmus paper and consequently contained a vigorous nitrifying flora which transformed the ammonia rapidly, even without the addition of lime.

In the presence of calcium oxide and acid phosphate, ammonium sulfate showed approximately one-third greater nitrate accumulation in the very

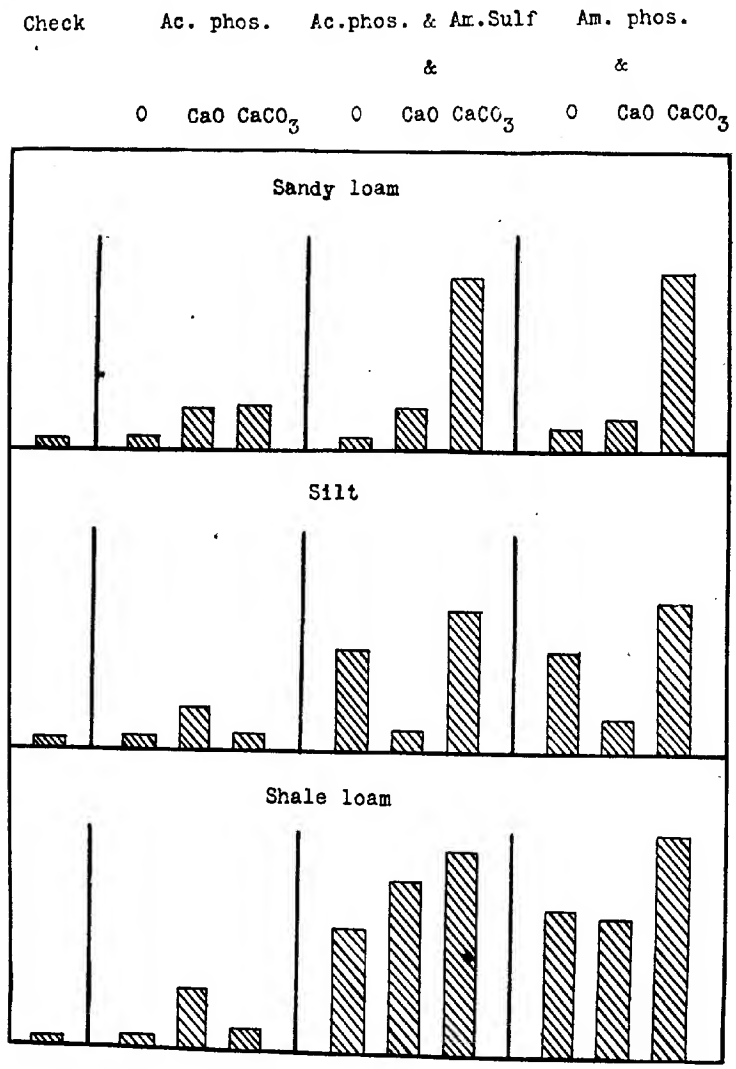

FIG. 4. DIAGRAM SHOWING THE EFFECT OF DIFFERENT FORMS OF LIME ON THE RATE OF NITRIFICATION OF AMMONIUM PHOSPHATE AND AMMONIUM SULFATE

acid sandy loam than did ammonium phosphate. In the shale loam there was also a difference of about 23 per cent in favor of ammonium sulfate where calcium oxide was used. However, the reverse is true in the silt loam, ammonium phosphate showing a 50 per cent increase over ammonium sulfate. It is interesting to note that the calcium oxide caused only comparatively slight increases in nitrate production in two soils and an actual depression in the third. Evidently calcium oxide is sufficiently caustic to kill some of the soil organisms, or at least to inhibit their proper multiplication in many cases.

Without exception the soils receiving calcium carbonate showed a greater accumulation of nitrates from both the ammonium fertilizers than any of the other treatments. It happened that in all three soils ammonium phosphate in the presence of calcium carbonate showed a slightly higher availability by nitrification than did ammonium sulfate, but the differences are nearly if not completely within experimental error.

In general, then, from the results reported in table 5 we may say that ammonium phosphate is directly comparable with ammonium sulfate in nitrifiability where acid phosphate is added to the ammonium sulfate in amounts equivalent to the phosphorous in ammonium phosphate. The three soils used were of quite widely different types, yet the fertilizers under a given set of conditions gave similar results.

Effect of ammonium phosphate upon ammonification

In order to determine if the addition of a fertilizer, like ammonium phosphate, which contains all of its nitrogen in the form of ammonia, would hinder the ammonification of organic matter added to the soil, the experiment recorded in table 6 was carried out. The soil chosen was a neutral sandy loam from the college farm, 100-gm. samples being weighed out into tumblers and the dried blood, green alfalfa and cottonseed meal thoroughly mixed with the sample according to the method followed in the nitrification experiments. Dried blood was applied at the rate of 2 gm. per tumbler, which expressed as nitrogen amounts to 252 mgm. The cottonseed meal and green alfalfa were analyzed and sufficient added to equal the nitrogen in the dried blood. After incubation at room temperature for 8 days the ammonia which had accumulated was determined by distillation with magnesium oxide. These results are given in table 6 and figure 5.

A study of the ammonification results shows that, in general, as the quantity of ammonium phosphate increases the amount of ammonia produced from dried blood decreases. The reverse is true of cottonseed meal, while the rate of ammonification of alfalfa remains practically constant. The large amount of ammonia produced from green alfalfa is especially significant, showing how rapidly the material will decay under favorable conditions. The depression of ammonification of dried blood where ammonium phosphate is used as contrasted with the increase with cottonseed meal is to be expected,

TABLE 6

Effect of ammonium phosphate upon ammonification

TREATMENT	NH_3 NITROGEN	AVERAGE	INCREASE OVER CHECK	PER CENT AMMONIFIED
	mgm.	*mgm.*	*mgm.*	
Check	1.69 1.47	1.58		
Dried blood	85.80 86.24	86.02	85.44	33.90
Cottonseed meal	66.79 67.75	67.27	65.69	26.07
Alfalfa	64.97 58.86	61.91	60.33	23.94
20 mgm. ammonium phosphate	1.47 1.84	1.65		
20 mgm. ammonium phosphate and dried blood	83.88 82.83	83.35	81.70	32.42
20 mgm. ammonium phosphate and cottonseed meal	71.89 73.97	72.93	71.28	28.28
20 mgm. ammonium phosphate and alfalfa	64.20 60.38	62.29	60.64	24.06
40 mgm. ammonium phosphate	2.84 3.03	2.93		
40 mgm. ammonium phosphate and dried blood	77.71 79.90	78.80	75.87	30.11
40 mgm. ammonium phosphate and cottonseed meal	71.69 73.10	72.39	69.46	27.56
40 mgm. ammonium phosphate and alfalfa	65.85 58.04	61.94	59.01	23.41
80 mgm. ammonium phosphate	7.61 6.42	7.01		
80 mgm. ammonium phosphate and dried blood	89.15 87.46	88.30	81.29	32.26
80 mgm. ammonium phosphate and cottonseed meal	80.79 85.27	83.03	76.02	30.16
80 mgm. ammonium phosphate and alfalfa	69.13 66.37	67.75	60.74	24.10
120 mgm. ammonium phosphate	11.64 11.74	11.69		
120 mgm. ammonium phosphate and dried blood	lost 91.19	91.19	79.50	31.15
120 mgm. ammonium phosphate and cottonseed meal	84.57 86.22	85.39	73.70	29.24

TABLE 6 (Concluded)

TREATMENT	NH_3 NITROGEN	AVERAGE	INCREASE OVER CHECK	PER CENT AMMONIFIED
	mgm.	*mgm.*	*mgm.*	
120 mgm. ammonium phosphate and alfalfa	69.48 73.31	71.39	59.70	23.69
160 mgm. ammonium phosphate	16.60 16.80	16.70		
160 mgm. ammonium phosphate and dried blood	87.56 95.62	91.59	74.89	29.71
160 mgm. ammonium phosphate and cottonseed meal	95.37 100.30	97.83	81.13	32.19
160 mgm. ammonium phosphate and alfalfa	76.69 78.06	77.37	60.67	24.06
200 mgm. ammonium phosphate	20.78 21.47	21.12		
200 mgm. ammonium phosphate and dried blood	99.19 93.07	96.13	75.01	29.76
200 mgm. ammonium phosphate and cottonseed meal	97.16 93.89	95.52	74.40	29.52
200 mgm. ammonium phosphate and alfalfa	82.05 78.80	80.42	59.30	23.53
300 mgm. ammonium phosphate	32.67 32.03	32.35		
300 mgm. ammonium phosphate and dried blood	105.35 103.66	104.50	72.15	28.63
300 mgm. ammonium phosphate and cottonseed meal	115.15 122.02	128.58	96.23	38.18
300 mgm. ammonium phosphate and alfalfa	100.74 93.53	97.13	64.78	25.71
1000 mgm. ammonium phosphate	105.25 105.05	105.15		
1000 mgm. ammonium phosphate and dried blood	163.38 173.13	168.25	63.10	25.04
1000 mgm. ammonium phosphate and cottonseed meal	204.27 192.04	198.15	93.00	36.91
1000 mgm. ammonium phosphate and alfalfa	169.95 159.42	164.67	59.52	23.62

since ammonium salts, and especially ammonium phosphate, stimulate the growth of fungi, which in turn can decompose cottonseed meal more rapidly than dried blood because of its wider nitrogen-carbon ratio.

Other investigators have reported similar results, among them being Lipman and Brown (18), who observed that sodium nitrate favors the growth of decay

bacteria. They attributed this to the fact that many saprophytic bacteria use nitrate and its presence, by increasing their numbers and vigor, results in an increased destruction of the humus. On the other hand, ammonium salts had a depressing effect upon the growth of bacteria but favored the fungi. The work here reported with ammonium phosphate, in general, gave similar results and agrees with the conclusions of these investigators concerning ammonium sulfate.

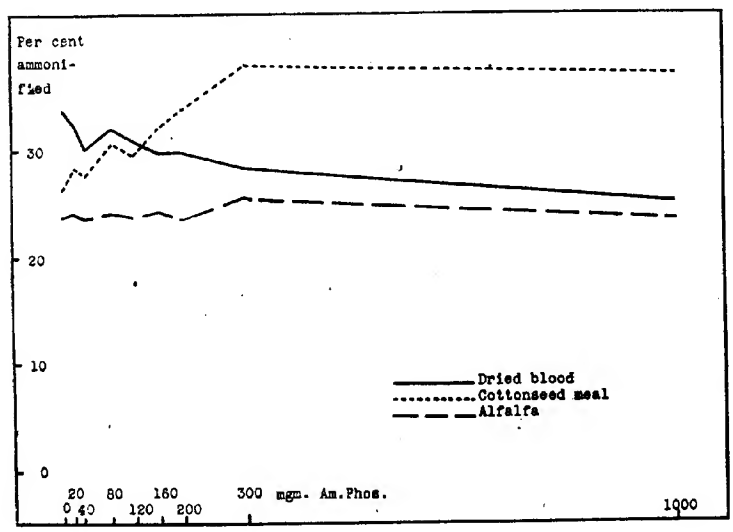

FIG. 5. DIAGRAM SHOWING THE EFFECT OF AMMONIUM PHOSPHATE UPON AMMONIFICATION

Utilization of ammonium phosphate and other nitrogenous salts by soil fungi

As a supplement to the ammonification and nitrification work, some experiments were started to determine just how readily ammonium phosphate is utilized by soil fungi. A large number of organisms were isolated directly from the field and a few of the most vigorous growers selected for this work. In the first series pure tri-basic ammonium phosphate was used for comparison with ammonium sulfate. Almost without exception the growth obtained with the six organisms used was greater in the flasks containing the ammonium phosphate. A later series comparing ammonium phosphate with ammonium sulfate, ammonium nitrate, urea, ammonium carbonate, sodium nitrate, and a mixture of ammonium sulfate and sodium nitrite, five pure cultures of soil fungi, being used, again showed ammonium phosphate to be more favorable to the organisms than ammonium sulfate. However, the ammonium carbonate medium was nearly as good as the ammonium phosphate. At the end

of the thirteenth day the ranking, as far as growth was concerned, was as follows:

First	Ammonium phosphate
Second	Ammonium carbonate
Third	Ammonium sulfate
Fourth	Urea
Fifth	Ammonium nitrate
Sixth	Sodium nitrate
Seventh	Ammonium sulfate and sodium nitrite (no growth in most cases)

One of the organisms used gave very poor growth in all cases except where the nitrogen supplied was in the form of ammonium phosphate.

The fact that soil bacteria and fungi can utilize ammonium phosphate as well as and often better than any other ammonium salt may or may not be a point in favor of the phosphate. Since the phosphate favors the growth of the organisms of the soil it necessarily means that more of the nitrogen will be converted into insoluble protein when it becomes a part of the organism itself; thus the ammonia or nitrate produced from it is taken away from the use of the plant. The nitrogen fixed in this manner will again be made available later when the organisms die and undergo decomposition. The rate at which these processes may occur in the soil is not well enough known to justify a definite statement as to the probability of the growth of the micro-organisms interfering with proper plant nourishment and development. Under normal conditions it is reasonable to suppose that the increased growth of bacteria and fungi following the application of ammonium phosphate would also act favorably on plant growth. Increased bacterial action causes more insoluble plant-food in the soil to become available and also hastens the decay of organic matter. The storing up of the nitrogen in the bodies of the soil organisms would merely fix it for the present and prevent a big stimulation of plant growth at the very first, followed by a decrease later, as often occurs when sodium nitrate is applied. There is still another factor to be considered. Under field conditions fertilizers are most commonly applied at about the time of planting. It is a month or longer, depending upon the crop being grown, before the plants will require very much nitrogen. The stimulation of bacteria and fungi, then, will result in the fixation of the nitrogen applied more quickly. In general, any change in the condition of the soil that has a beneficial effect on the bacteria in the soil will act favorably on the plants also.

Pot experiments

The purpose of these experiments was to determine the relative availability of the nitrogen and of the phosphorous in the ammonium phosphate fertilizer under varying soil conditions and with different soils. The effect of the common fertilizers on soil reaction also was among the points considered, especially in relation to the availability of each of the fertilizers. The ques-

tions, does ammonium phosphate show a residual effect and how is the soil affected by long continued use, also are at least partially answered.

Comparative availability of various nitrogenous fertilizers and their effect upon soil reaction under greenhouse conditions

For this experiment two soils were selected, a Norfolk sand and a Sassafras loam. The first of these was light in texture, low in fertility and had a lime-requirement of 714 pounds of calcium carbonate per 3,000,000 pounds of soil. The second was low in crop production, also, probably because of acidity rather than lack of nitrogen, phosphorous or potassium. Its lime-requirement was found to be 5000 pounds of calcium carbonate per acre. The soils were air-dried, sieved and weighed out into pots holding 20 pounds of soil. The fertilizers applied in addition to ammonium phosphate were ammonium sulfate, dried blood, and cottonseed meal. Two rates of application were made, namely, 1.5 gm. and 3 gm. of ammonium phosphate per pot and equivalent amounts of nitrogen in the case of the other three fertilizers. Half of the pots received lime and the other half were left unlimed. In all cases where lime was applied enough was used to neutralize the acidity and 2 tons of calcium carbonate in excess. Two grams of potassium chloride were added to all pots and enough 16 per cent acid phosphate was applied to those pots not receiving ammonium phosphate to equal the phosphorous in this fertilizer. This was more phosphorous than the plants actually required but it was necessary to use this amount in order to eliminate the element as a variable factor. As is brought out later in the germination experiments, very large quantities of acid phosphate are required to injure plants and therefore the excess would not be expected to cause an injury. The fertilizers were mixed with the entire 20 pounds of soil, and barley planted on December 31, 1915. Soon after the plants were up mildew attacked them so badly that the crop was harvested on February 10 when the plants were about 10 inches in height. The plants were dried and analyzed and the results reported in table 7.

No importance should be attached to these results since in many cases the fungus greatly decreased the yields. Immediately after harvesting the barley the pots were planted to buckwheat. This crop was chosen because of its quick growth and adaptability to greenhouse conditions and, above all, because it is not attacked by the downy mildew. The yields and analyses are given in table 8.

After removing the crop the pots were again planted to buckwheat without the use of additional fertilizer. The results are given in table 9. As will be noticed from the yields of the last crop the available plant-food had been practically exhausted by the barley and the two residual crops of buckwheat. The total yields of dry matter and the recoveries of nitrogen from these three crops are summarized in table 10.

A glance at the summary table gives a fair indication of the availability

TABLE 7

Comparative availability of various nitrogenous fertilizers in soil under greenhouse conditions
First crop (barley)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
	Norfolk sand (167 mgm. of nitrogen added)								
Unlimed									
		grams	*grams*	*gram*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
1	Check	1.878			2.77		52.0		
2		1.707	1.793		3.17	2.97	54.1	53.05	
3	Ammonium phosphate	2.406			4.22		101.5		
4		2.278	2.342	0.549	3.90	4.06	88.9	95.20	42.15
5	Ammonium sulfate	2.342			4.01		94.0		
6		2.356	2.349	0.556	4.01	4.01	94.4	94.20	41.15
7	Dried blood	2.192			3.55		77.7		
8		2.418	2.305	0.512	3.52	3.54	85.2	81.45	28.40
9	Cottonseed meal	2.269			3.53		80.0		
10		2.255	2.262	0.469	3.60	3.57	81.1	80.55	27.50
Limed									
11	Check	2.023			3.24		65.6		
12		1.713	1.868		3.53	3.39	60.4	63.00	
13	Ammonium phosphate	2.013			3.78		76.0		
14		2.127	2.070	0.202	3.78	3.78	80.3	78.15	15.15
15	Ammonium sulfate	1.768			3.89		68.7		
16		2.007	1.888	0.020	3.67	3.78	73.7	71.20	8.20
17	Dried blood	2.143			3.50		75.0		
18		1.810	1.977	0.109	3.77	3.64	68.2	71.60	8.60
19	Cottonseed meal	2.079			3.81		79.2		
20		1.965	2.022	0.154	3.64	3.73	71.6	75.40	12.40
	Sassafras loam (167 mgm. of nitrogen added)								
Unlimed									
21	Check	2.304			4.42		101.80		
22		2.805	2.555		4.18	4.30	117.26	109.53	
23	Ammonium phosphate	2.377			4.33		102.98		
24		1.958	2.168	0.387	4.47	4.40	87.60	95.29	−14.24
25	Ammonium sulfate	2.458			4.48		108.34		
26		2.176	2.317	−0.238	4.63	4.56	100.70	104.52	− 5.01
27	Dried blood	2.130			4.35		92.70		
28		2.636	2.383	−0.172	4.35	4.35	114.60	103.65	− 5.88
29	Cottonseed meal	2.301			4.41		101.40		
30		2.091	2.196	−0.359	4.51	4.46	94.30	97.85	−11.68

TABLE 7—(Continued)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
Limed									
		grams	*grams*	*gram*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
31	Check	2.283			4.52		103.15		
32		2.721	2.452		4.79	4.66	130.45	116.80	
33	Ammonium phosphate	2.096			5.10		106.80		
34		2.471	2.283	−0.169	4.47	4.79	110.55	108.67	− 8.13
35	Ammonium sulfate	1.954			5.28		103.20		
36		2.264	2.109	−0.343	4.61	4.95	105.75	104.47	−12.33
37	Dried blood	2.206			4.96		109.48		
38		2.368	2.287	−0.165	4.61	4.79	108.90	109.19	− 7.61
39	Cottonseed meal	2.071			4.51		93.45		
40		2.341	2.206	−0.246	4.25	4.38	99.55	96.47	−20.33
Norfolk sand (334 mgm. of nitrogen added)									
Unlimed									
41	Check	1.927			3.12		60.20		
42		2.028	1.978		3.08	3.10	62.40	61.30	
43	Ammonium phosphate	2.941			3.98		117.05		
44		2.363	2.656	0.678	4.03	4.01	95.25	106.15	44.85
45	Ammonium sulfate	2.354			4.10		96.60		
46		2.919	2.637	0.659	4.28	4.19	125.00	110.80	49.50
47	Dried blood	2.709			3.87		104.80		
48		2.265	2.487	0.509	3.87	3.87	87.70	96.25	34.95
49	Cottonseed meal	2.656			3.98		105.80		
50		2.697	2.677	0.699	3.94	3.96	106.15	105.97	44.67
Limed									
51	Checked	1.883			4.28		80.70		
52		2.102	1.993		3.27	3.78	68.80	74.75	
53	Ammonium phosphate	2.321			3.82		88.10		
54		2.565	2.443	0.451	3.57	3.70	87.20	87.65	12.90
55	Ammonium sulfate	2.079			4.03		83.80		
56		2.532	2.306	0.313	3.93	3.98	99.45	91.62	16.87
57	Dried blood	2.318			3.94		91.25		
58		2.266	2.292	0.299	3.76	3.85	85.30	88.27	13.52
59	Cottonseed meal	2.552			3.73		95.10		
60		2.626	2.589	0.597	3.75	3.74	98.25	96.67	21.92
Sassafras loam (334 mgm. of nitrogen added)									
Unlimed									
61	Check	3.333			4.37		145.70		
62		3.745	3.539		4.35	4.36	162.80	154.25	
63	Ammonium phosphate	3.224			4.42		142.65		
64		3.123	3.177	−0.362	4.48	4.45	140.10	141.37	−12.88

TABLE 7—(Concluded)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
	Sassafras loam (334 mgm. of nitrogen added)								
Unlimed									
		grams	*grams*	*gram*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
65	Ammonium sulfate...	3.640			4.46		162.30		
66		3.436	3.538	−0.001	4.26	4.36	146.36	154.33	0.08
67	Dried blood.....	3.983			4.35		173.30		
68		4.224	4.104	0.565	4.79	4.57	202.20	187.75	33.50
69	Cottonseed meal.....	4.216			4.30		181.24		
70		4.579	4.398	0.859	4.17	4.24	191.24	186.24	31.99
Limed									
71	Check.....	4.406			4.34		191.42		
72		4.173	4.290		4.64	4.49	194.25	192.83	
73	Ammonium phosphate.....	3.485			4.61		160.68		
74		3.649	3.567	−0.723	4.56	4.59	166.35	163.51	−29.32
75	Ammonium sulfate...	3.732			4.55		169.76		
76		3.431	3.583	−0.707	4.62	4.59	158.65	164.20	−28.63
77	Dried blood.....	3.809			4.48		170.70		
78		3.748	3.779	−0.511	4.50	4.49	168.66	169.68	−23.15
79	Cottonseed meal.....	3.169			4.41		139.60		
80		3.656	3.413	−0.877	4.59	4.50	168.10	153.85	−38.98

of ammonium phosphate as compared with other nitrogenous fertilizers. While in most cases the comparative availability of any two fertilizers is very nearly the same whether judged from the standpoint of crop yield or the recovery of nitrogen, it is better in this particular case to use the recovery of nitrogen as a basis of comparison, since two different kinds of crops were grown.

In the case of pots 1 to 10, containing Norfolk sand without lime, ammonium phosphate ranked appreciably above ammonium sulfate and very much above the two organic fertilizers. When the same soil was limed the ratio of one fertilizer to another was practically the same, except that for some unknown reason cottonseed meal gave very much lower results.

Referring to pots 41 to 60, where the same experiment was repeated except that twice as much nitrogen was applied, it will be noticed that the same ratios hold true. However, ammonium sulfate gave yields more nearly equal to ammonium phosphate, especially in the unlimed soil. The highest percentage recovery of the nitrogen applied was 69.94 per cent.

As previously stated the Norfolk sand was very poor and only slightly acid, thus explaining why liming failed to cause any decided increases in crop yields. Considering the recovery of nitrogen in the Sassafras loam it will be noticed in the summary table that for pots 21 to 30, which were unlimed,

TABLE 8

Comparative availability of various nitrogenous fertilizers in soil under greenhouse conditions
Second crop (Buckwheat)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
				Norfolk sand					
Unlimed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
1	Check	4.6			1.29		59.34		
2		4.0	4.3		1.16	1.23	46.40	52.87	
3	Ammonium phosphate	15.4			0.73		112.42		
4		16.7	16.05	11.75	0.72	0.73	120.24	116.33	63.46
5	Ammonium sulfate	15.7			0.68		106.76		
6		14.4	15.05	10.75	0.73	0.71	105.12	105.94	63.97
7	Dried blood	11.2			0.83		92.96		
8		10.6	10.9	6.60	1.12	0.98	118.72	105.84	52.97
9	Cottonseed meal	10.5			0.83		87.15		
10		11.0	10.75	6.45	0.83	0.83	91.30	89.23	36.36
Limed									
11	Check	10.9			1.03		112.27		
12		10.6	10.75		0.92	0.98	97.52	104.90	
13	Ammonium phosphate	26.2			0.70		183.40		
14		22.5	24.35	13.60	0.80	0.75	180.00	181.70	76.80
15	Ammonium sulfate	20.0			0.86		162.00		
16		22.8	21.40	10.65	0.74	0.80	168.72	165.36	60.46
17	Dried blood	20.2			0.79		159.58		
18		20.6	20.40	9.65	0.81	0.80	166.86	163.22	58.32
19	Cottonseed meal	17.0			0.82		139.40		
20		15.7	16.35	5.60	0.86	0.84	135.02	137.21	32.31
				Sassafras loam					
Unlimed									
21	Check	24.8			0.99		245.52		
22		25.0	24.90		0.99	0.99	247.52	246.51	
23	Ammonium phosphate	26.3			1.15		302.45		
24		35.0	30.65	5.75	0.97	1.06	304.50	303.23	56.72
25	Ammonium sulfate	25.3			1.29		326.37		
26		27.0	26.15	1.25	1.36	1.32	367.20	346.79	100.75
27	Dried blood	29.0			1.11		321.90		
28		33.2	31.10	6.20	1.06	1.09	351.92	336.91	90.40
29	Cottonseed meal	30.2			1.12		338.24		
30		29.4	29.80	4.90	1.05	1.09	308.70	323.47	76.96

TABLE 8—(Continued)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
Limed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
31	Check	37.6			0.85		319.60		
32		32.3	34.95		0.87	0.86	281.01	300.31	
33	Ammonium phosphate	lost							
34		36.6	36.6	1.65	1.18	1.180	431.88	431.88	131.57
35	Ammonium sulfate	lost							
36		36.8	36.80	1.85	1.12	1.120	412.16	412.16	111.85
37	Dried blood	lost							
38		37.8	37.80	2.85	1.03	1.030	389.34	389.34	89.03
39	Cottonseed meal	30.0			1.01		303.00		
40		37.2	33.60	−1.35	0.96	0.985	357.12	330.06	29.75
				Norfolk sand					
Unlimed									
41	Check	6.8			1.01		68.68		
42		5.1	5.95		1.21	1.110	61.71	65.20	
43	Ammonium phosphate	35.5			0.63		223.65		
44		35.5	35.50	29.55	0.67	0.650	237.85	230.75	165.55
45	Ammonium sulfate	32.5			0.71		230.75		
46		34.8	33.65	27.70	0.62	0.665	215.76	223.26	158.56
47	Dried blood	24.3			0.70		170.10		
48		26.8	25.55	19.60	0.73	0.715	195.64	182.87	117.67
49	Cottonseed meal	21.5			0.73		156.95		
50		21.0	21.25	15.30	0.74	0.735	155.40	156.18	90.98
Limed									
51	Check	9.1			0.90		81.90		
52		10.3	9.70		0.87	0.89	89.61	85.76	
53	Ammonium phosphate	34.6			0.83		287.18		
54		33.8	34.20	24.50	0.83	0.83	280.54	283.86	198.10
55	Ammonium sulfate	33.4			0.82		273.88		
56		34.7	34.05	24.35	0.79	0.81	274.13	274.01	188.25
57	Dried blood	23.2			0.85		197.20		
58		31.6	27.40	17.70	0.83	0.84	265.44	231.32	145.56
59	Cottonseed meal	24.8			0.83		205.84		
60		20.0	22.40	12.70	0.87	0.85	174.00	189.92	104.16

TABLE 8—(Concluded)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
				Sassafras loam					
Unlimed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
61	Check	21.5			1.02		219.30		
62		17.6	19.55		1.33	1.18	234.08	226.69	
63	Ammonium phosphate	36.5			1.18		430.70		
64		36.6	36.55	17.00	1.19	1.19	435.54	433.12	206.43
65	Ammonium sulfate	21.5			2.09		449.50		
66		23.0	22.25	3.70	2.19	2.14	503.70	476.60	249.91
67	Dried blood	28.2							
68		lost	28.20	8.65	1.41	1.41	397.62	397.62	170.93
69	Cottonseed meal	25.8			1.18		304.44		
70		28.6	27.20	7.65	1.17	1.18	334.62	319.53	92.84
Limed									
71	Check	28.6			1.04		297.44		
72		38.4	33.50		1.08	1.06	414.72	356.08	
73	Ammonium phosphate	43.8			1.07		468.66		
74		40.5	42.15	8.65	1.07	1.07	433.35	451.01	94.93
75	Ammonium sulfate	44.5			1.10		489.50		
76		45.5	45.00	11.50	1.03	1.07	468.65	479.08	123.00
77	Dried blood	35.6			1.08		384.48		
78		40.8	38.20	4.70	1.02	1.05	416.16	400.32	44.24
79	Cottonseed meal	36.4			1.05		382.20		
80		38.7	37.55	4.05	1.02	1.04	394.74	388.47	32.39

ammonium sulfate ranked first, ammonium phosphate second, and dried blood and cottonseed meal, third and fourth, respectively. When lime, was used, the recovery from ammonium phosphate was nearly 7 per cent higher than from ammonium sulfate and decidedly above that from the two organic fertilizers.

In pots 61 to 80 where large quantities of nitrogen were used similar results were obtained, with the exception that ammonium phosphate stood second in both cases. This Sassafras loam which was not very much heavier than the Norfolk sand was more acid and richer in nitrogen. This explains the results obtained, especially as regards the low recovery of nitrogen in most cases. Liming made the nitrogen already in the soil available, so that fertilizers did not always show large increases. It is to be regretted that "damping-off" fungi attacked the young plants in a few of the pots and results from these cannot be given. Pots 33, 35 and 37 accidently received an application of fertilizer after the crop of barley was grown and these must be eliminated in considering the data.

TABLE 9

Comparative availability of various nitrogenous fertilizers in soil under greenhouse conditions. Third crop (buckwheat)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
				Norfolk sand					
Unlimed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
1	Check	0.0			0.00		0.00		
2		0.1	0.05		2.10	2.10	2.10	1.05	
3	Ammonium phosphate	0.1			2.10		2.10		
4		0.6	0.35	0.30	1.85	1.98	10.90	6.50	5.45
5	Ammonium sulfate	0.1			2.05		2.05		
6		0.0	0.05	0.00	0.00	2.05	0.00	1.02	−0.03
7	Dried blood	0.6			1.75		10.50		
8		0.1	0.35	0.30	2.00	1.88	2.00	6.25	5.20
9	Cottonseed meal	0.6			1.99		11.90		
10		0.3	0.45	0.40	1.77	1.88	5.30	8.55	7.50
Limed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
11	Check	0.6			1.85		11.10		
12		1.0	0.80		1.71	1.78	17.10	14.10	
13	Ammonium phosphate	1.9			1.66		31.54		
14		1.4	1.65	0.85	1.75	1.71	24.50	28.02	13.92
15	Ammonium sulfate	2.7			1.54		41.58		
16		1.2	1.95	1.15	1.51	1.53	18.12	29.85	15.75
17	Dried blood	2.0			1.59		31.80		
18		1.8	1.90	1.10	1.34	1.47	24.12	27.96	13.86
19	Cottonseed meal	0.4			1.85		7.40		
20		1.3	0.85	0.05	1.14	1.50	14.82	11.11	−2.99
				Sassafras loam					
Unlimed									
21	Check	2.4			1.41		33.84		
22		2.0	2.20		1.74	1.58	34.80	34.32	
23	Ammonium phosphate	2.9			1.88		54.52		
24		1.9	2.40	−0.20	1.69	1.79	32.11	43.32	9.00
25	Ammonium sulfate	1.2			2.00		24.00		
26		2.5	1.85	−0.35	1.55	1.78	38.75	31.38	−2.94
27	Dried blood	1.0			2.36		23.60		
28		2.1	1.55	−0.65	1.67	2.12	35.07	29.34	−4.98
29	Cottonseed meal	2.0			1.73		34.60		
30		1.9	1.95	−0.25	1.91	1.82	36.29	35.45	1.13

TABLE 9—(Continued)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
				Sassafras loam					
Limed									
31	Check	2.5			1.89		47.25		
32		4.4	3.45		1.59	1.74	69.96	58.61	
33	Ammonium phosphate	lost							
34		3.1	3.10	−0.35	1.73	1.73	53.63	53.63	−4.98
35	Ammonium sulfate	lost							
36		4.0	4.00	0.55	1.67	1.67	66.80	66.80	8.19
37	Dried blood	lost							
38		4.3	4.30	0.85	1.53	1.53	65.79	65.79	7.18
39	Cottonseed meal	5.3			1.73		91.69		
40		3.0	4.15	0.70	1.87	1.80	56.10	73.90	15.29
				Norfolk sand					
Unlimed		*grams*	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
41	Check	0.0			0.00		0.00		
42		0.0	0.00		0.00	0.00	0.00	0.00	
43	Ammonium phosphate	0.3			2.58		7.74		
44		0.2	0.25	0.25	2.62	2.60	5.24	6.49	6.49
45	Ammonium sulfate	0.2			2.62		5.24		
46		0.1	0.15	0.15	2.50	2.56	2.50	3.87	3.87
47	Dried blood	0.4			2.33		9.32		
48		0.6	0.50	0.50	1.68	2.01	10.08	9.70	9.70
49	Cottonseed meal	0.1			2.05		2.05		
50		0.9	0.50	0.50	2.11	2.08	18.99	10.52	10.52
Limed									
51	Check	0.5			2.41		11.05		
52		0.0	0.25		0.00	2.41	0.00	5.53	
53	Ammonium phosphate	1.2			2.21		26.52		
54		1.7	1.45	1.20	1.75	1.98	29.75	28.14	22.61
55	Ammonium sulfate	0.8			2.01		16.08		
56		0.2	0.50	0.25	1.91	1.96	3.82	9.95	4.42
57	Dried blood	1.6			1.98		31.68		
58		1.9	1.75	1.50	2.73	2.36	51.87	41.78	36.25
59	Cottonseed meal	0.0		0.00	0.00		0.00		
60		0.7	0.35	0.10	2.01	2.01	14.07	7.04	1.51

TABLE 9—(Concluded)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
				Sassafras loam					
Unlimed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
61	Check	0.5			2.39		11.95		
62		2.0	1.25		1.77	2.08	35.40	23.68	
63	Ammonium phosphate	2.0			1.90		38.00		
64		2.0	2.00	0.75	2.31	2.11	46.20	42.10	18.42
65	Ammonium sulfate	2.5			2.36		59.00		
66		2.3	2.40	1.15	2.17	2.27	49.91	54.46	30.78
67	Dried blood	2.5			2.05		51.25		
68		lost	2.50	1.25		2.05		51.25	27.57
69	Cottonseed meal	2.6			1.89		49.14		
70		1.5	2.05	0.80	2.73	2.31	40.95	45.07	21.39
Limed									
71	Check	2.7			2.13		57.51		
72		5.5	4.10		1.81	1.97	99.55	78.53	
73	Ammonium phosphate	3.2			1.79		57.28		
74		5.0	4.10	0.00	1.75	1.77	87.50	72.39	−6.14
75	Ammonium sulfate	2.6			1.84		47.84		
76		3.6	3.10	−1.00	1.57	1.71	56.52	52.18	−26.15
77	Dried blood	4.2			1.70		61.40		
78		3.6	3.90	−0.20	1.85	1.78	66.60	64.00	−14.53
79	Cottonseed meal	5.3			1.76		93.28		
80		4.5	4.90	0.80	1.80	1.78	81.00	87.14	8.61

One peculiarity is brought out in the results, namely, the low crop yield in the unlimed ammonium sulfate pots as compared with ammonium phosphate, but the very much higher percentage of nitrogen in the crop. It is barely possible that under the existing acid conditions the nitrogen of ammonium sulfate is less efficient as far as increasing crop growth is concerned, but the writer does not wish to state this definitely.

From the results as a whole one might say on first thought that ammonium phosphate would give better results if applied on light sandy soils while the reverse would be true for ammonium sulfate, but the data given do not prove this point. Undoubtedly the chemical and biological differences between the two soils are much greater factors than the mere textural differences. Other sandy or loam soils would probably give entirely different results.

As has been previously shown by various investigators dried blood and cottonseed meal rank much lower in availability than ammonium sulfate. In both soils used in this experiment dried blood showed a tendency to produce larger yields than cottonseed meal. The residual effect which is often claimed

TABLE 10

Comparative availability of various nitrogenous fertilizers in soil under greenhouse conditions
Summary—First three crops

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	TOTAL NITROGEN RECOVERED	AVERAGE	INCREASE OVER CHECK	PER CENT RECOVERED
		Norfolk sand (167 mgm. of nitrogen added)						
Unlimed								
		grams	*grams*	*grams*	*mgm.*	*mgm.*	*mgm.*	
1	Check	6.49			111.34			
2		5.81	6.15		102.60	106.97		
3	Ammonium phosphate	17.91			216.02			
4		19.58	18.75	12.60	220.04	218.03	111.06	66.50
5	Ammonium sulfate	18.14			202.86			
6		16.76	17.45	11.30	199.52	201.19	94.22	56.41
7	Dried blood	13.99			181.16			
8		13.12	13.56	7.41	202.17	191.67	84.70	50.72
9	Cottonseed meal	13.37			179.05			
10		13.56	13.47	7.32	177.70	178.38	71.41	42.76
Limed								
11	Check	13.52			188.97			
12		13.31	13.42		172.02	180.50		
13	Ammonium phosphate	30.11			290.94			
14		26.03	28.07	14.65	284.80	287.87	107.37	64.29
15	Ammonium sulfate	24 47			272.28			
16		26.01	25.24	11.82	260.54	266.41	85.91	51.44
17	Dried blood	24.34			266.38			
18		24.21	24.28	10.86	259.18	262.78	82.28	49.27
19	Cottonseed meal	19.48			226.00			
20		18.97	19.23	5.81	221.44	223.72	43.22	25.88
		Sassafras loam (167 mgm. of nitrogen added)						
Unlimed								
21	Check	29.50			381.16			
22		29.81	29.66		399.56	390.36		
23	Ammonium phosphate	31.58			494.08			
24		38.86	35.22	5.56	459.21	476.65	86.29	51.57
25	Ammonium sulfate	28.96			458.71			
26		31.68	30.32	0.66	506.65	482.68	92.32	55.28
27	Dried blood	32.13			438.20			
28		37.94	35.04	5.38	501.59	469.90	79.54	47.63
29	Cottonseed meal	34.50			474.24			
30		33.39	33.95	4.29	439.29	456.74	66.38	39.75

TABLE 10—(Continued)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	TOTAL NITROGEN RECOVERED	AVERAGE	INCREASE OVER CHECK	PER CENT RECOVERED
		Sassafras loam (167 mgm. of nitrogen added)						
Limed								
		grams	*grams*	*grams*	*mgm.*	*mgm.*	*mgm.*	
31	Check	42.38			470.00			
32		39.42	40.90		481.42	475.71		
33	Ammonium phosphate	lost						
34		42.17	42.17	1.27	596.06	596.06	120.35	72.06
35	Ammonium sulfate	lost						
36		43.06	43.06	2.16	584.11	584.11	109.00	65.27
37	Dried blood	lost						
38		44.47	44.47	3.57	564.03	564.03	88.32	52.89
39	Cottonseed meal	37.37			488.14			
40		42.54	39.96	−0.94	512.77	500.46	24.75	14.82
		Norfolk sand (334 mgm. of nitrogen added)						
Unlimed								
41	Check	8.73			128.88			
42		7.13	7.93		124.11	126.50		
43	Ammonium phosphate	38.74			348.44			
44		38.06	38.40	30.47	338.34	343.39	216.89	64.87
45	Ammonium sulfate	35.05			332.59			
46		37.82	36.44	28.51	343.26	337.93	209.44	62.71
47	Dried blood	27.41			284.22			
48		29.67	28.54	20.61	293.42	288.82	162.32	48.60
49	Cottonseed meal	24.26			264.80			
50		24.60	24.43	16.50	280.54	272.67	146.17	43.76
Limed								
51	Check	11.48			173.65			
52		12.40	11.99		158.41	166.03		
53	Ammonium phosphate	38.12			401.80			
54		38.07	38.10	26.11	397.49	399.65	233.62	69.94
55	Ammonium sulphate	36.28			373.76			
56		37.43	36.86	24.87	377.40	375.58	209.55	62.73
57	Dried blood	27.12			320.13			
58		35.77	31.45	19.46	302.61	311.37	145.34	43.51
59	Cottonseed meal	27.35			300.94			
60		23.33	25.34	13.35	286.32	296.63	127.60	38.20

TABLE 10—(Concluded)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	TOTAL NITROGEN RECOVERED	AVERAGE	INCREASE OVER CHECK	PER CENT RECOVERED
	Sassafras loam (334 mgm. of nitrogen added)							
Unlimed								
		grams	*grams*	*grams*	*mgm.*	*mgm.*	*mgm.*	
61	Check	25.33			376.95			
62		23.35	24.34		432.28	404.62		
63	Ammonium phosphate	41.72			611.35			
64		41.72	41.72	17.38	621.84	616.60	211.98	63.46
65	Ammonium sulfate	27.64			670.80			
66		28.74	28.19	3.85	699.97	685.39	280.77	84.06
67	Dried blood	34.68						
68		lost	34.68	10.34	622.17	622.17	217.55	65.13
69	Cottonseed meal	32.62			534.82			
70		34.68	33.65	9.31	566.81	550.82	146.20	43.77
Limed								
71	Check	35.71			546.37			
72		48.07	41.89		708.52	627.45		
73	Ammonium phosphate	50.49			686.62			
74		49.15	49.82	7.93	687.20	686.91	59.46	17.80
75	Ammonium sulfate	50.83			707.09			
76		52.53	51.68	9.79	683.82	695.46	68.01	20.36
77	Dried blood	43.61			616.58			
78		48.15	45.88	3.99	651.42	634.00	6.55	1.96
79	Cottonseed meal	44.87			615.08			
80		46.86	45.87	3.98	643.84	629.46	2.01	0.60

for the organic fertilizers did not seem to be very marked under greenhouse conditions. The third crop in many cases is just as large in the ammonium phosphate or ammonium sulfate pots as in the others.

After removing the three crops, the yields and analyses of which were given in previous tables, the fertilizer applications used at the beginning of the experiment were repeated except that no lime was added. The soils were removed from the pots and the fertilizers mixed with the entire 20 pounds of soil. The yields of the buckwheat and analyses are given in table 11. A residual crop of buckwheat was also grown and the data reported in table 12. Where the crop yield was less than one gram the sample was not analyzed but the proportion of nitrogen was assumed to be 1.7 per cent. The slight error introduced is practically negligible. The buckwheat plants in a few more pots were destroyed by "damping-off" fungi and the yields are not given. The residual crop of buckwheat was so small that it was not considered worth while to plant another crop without additional fertilizer, as was done in the

TABLE 11

Comparative availability of various nitrogenous fertilizers in soil under greenhouse conditions
Fourth crop (buckwheat)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
	Norfolk sand (167 mgm. of nitrogen added)								
Unlimed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
1	Check	3.5			1.41		49.35		
2		1.5	2.50		1.56	1.48	23.40	36.37	
3	Ammonium phosphate	16.2			1.05		170.10		
4		15.7	15.95	13.45	0.98	1.01	153.86	161.98	125.61
5	Ammonium sulfate	13.7			1.14		156.18		
6		15.4	14.55	12.05	1.36	1.25	209.44	182.81	146.44
7	Dried blood	9.0			1.29		116.10		
8		8.4	8.70	6.20	1.21	1.25	101.64	108.87	72.50
9	Cottonsed meal	10.5			1.09		114.45		
10		10.5	10.50	8.00	0.92	1.00	96.60	105.52	69.15
Limed									
11	Check	5.0			1.35		67.50		
12		4.6	4.80		1.24	1.29	57.04	62.27	
13	Ammonium phosphate	18.1			1.02		184.62		
14		16.7	7.40	12.60	1.13	1.07	188.71	186.66	124.39
15	Ammonium sulfate	16.0			1.07		171.20		
16		16.7	16.35	11.55	1.07	1.07	178.69	174.94	112.67
17	Dried blood	11.7			1.04		121.68		
18		13.9	12.80	8.00	1.07	1.05	148.73	135.20	72.93
19	Cottonseed meal	13.7			0.99		135.63		
20		11.2	12.45	7.65	1.15	1.07	128.80	132.21	69.94
	Sassafras loam (167 mgm. of nitrogen added)								
Unlimed									
21	Check	1.1			2.26		24.86		
22		1.2	1.15		2.05	2.15	24.60	24.73	
23	Ammonium phosphate	5.4			2.23		120.42		
24		4.0	4.70	3.55	2.35	2.29	94.00	107.21	82.48
25	Ammonium sulfate	4.1			2.60		106.60		
26		2.2	3.15	2.00	2.52	2.56	55.44	81.02	57.29
27	Dried blood	2.6			2.69		69.94		
28		4.6	3.60	2.55	2.33	2.51	107.18	88.56	63.83
29	Cottonseed meal	3.2			2.29		73.28		
30		3.0	3.10	1.95	2.41	2.35	72.30	72.79	48.06

TABLE 11—(Continued)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
	Sassafras loam (167 mgm. of nitrogen added)								
Limed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
31	Check	8.0			1.20		96.00		
32		8.7	8.35		1.12	1.16	97.44	96.72	
33	Ammonium phosphate	lost							
34		lost	lost						
35	Ammonium sulfate	lost							
36		18.0	18.00	9.65	1.24	1.24	223.20	223.20	126.48
37	Dried blood	lost							
38		13.7	13.70	5.35	1.26	1.26	172.62	172.62	75.90
39	Cottonseed meal	15.2			1.21		183.92		
40		16.5	15.85	7.50	1.15	1.18	189.75	186.83	90.11
	Norfolk sand (334 mgm. of nitrogen added)								
Unlimed									
41	Check	0.8			1.79		14.32		
42		0.0	0.40		0.00	1.79	0.00	7.16	
43	Ammonium phosphate	23.1			1.15		265.65		
44		22.7	22.90	22.50	1.20	1.17	272.40	269.02	262.86
45	Ammonium sulfate	14.2			1.84		261.28		
46		15.2	14.70	14.30	1.67	1.75	253.84	257.56	250.40
47	Dried blood	11.8			1.32		155.76		
48		12.9	12.36	11.95	1.31	1.31	168.99	162.37	155.21
49	Cottonseed meal	16.8			1.19		199.92		
50		14.8	15.80	15.40	1.31	1.25	193.88	196.90	189.74
Limed									
51	Check	3.3			1.31		43.23		
52		4.6	3.90		1.37	1.34	61.65	52.44	
53	Ammonium phosphate	29.5							
54		lost	29.50	25.60	1.13	1.13	333.35	333.35	280.91
55	Ammonium sulfate	24.0			1.21		290.40		
56		25.3	24.65	20.75	1.15	1.18	290.95	290.67	238.23
57	Dried blood	17.6			1.36		239.36		
58		20.2	18.90	15.00	1.26	1.31	254.52	246.94	194.50
59	Cottonseed meal	19.6			1.20		235.20		
60		22.0	20.80	16.90	1.12	1.16	246.40	240.80	188.36

TABLE 11—(Concluded)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
	Sassafras loam (334 mgm. of nitrogen added)								
Unlimed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
61	Check	2.1			1.91		40.11		
62		0.6	1.35		2.01	1.96	12.06	26.08	
63	Ammonium phosphate	6.0			2.39		143.40		
64		5.6	5.80	4.45	2.62	2.50	146.72	145.06	118.98
65	Ammonium sulfate	4.2			2.55		107.10		
66		5.2	4.70	3.35	2.74	2.64	142.48	124.79	98.71
67	Dried blood	2.4			2.56		61.44		
68		2.9	2.65	1.30	3.29	2.92	95.41	78.42	52.34
69	Cottonseed meal	5.5			2.51		138.05		
70		5.6	5.55	4.20	2.41	2.46	134.96	136.50	110.42
Limed									
71	Check	7.5			1.24		93.00		
72		9.2	8.35		1.35	1.29	124.20	108.60	
73	Ammonium phosphate	lost							
74		23.5	23.50	15.15	1.27	1.27	298.45	298.45	189.85
75	Ammonium sulfate	26.6			1.26		335.16		
76		31.0	28.80	20.45	1.08	1.17	334.80	334.98	226.38
77	Dried blood	23.0			1.35		310.50		
78		22.4	22.70	14.35	1.27	1.31	284.48	297.49	188.89
79	Cottonseed meal	20.6			1.08		222.48		
80		23.9	22.25	13.90	1.22	1.15	291.58	257.03	148.43

first portion of this experiment. The summarized results for the two crops are given in table 13.

In the following discussion of the results obtained subsequent to the second application of fertilizers to the soil it should be borne in mind that the recovery of nitrogen is probably influenced to some extent, at least, by the previous application of fertilizers. Just to what extent this factor enters in cannot be stated but it is certainly true that in most cases the recoveries of nitrogen during the second stage of the experiment were greater than for the original application. This is in agreement with the work of Lipman and Blair (22) who found that in cylinder experiments the average recoveries of nitrogen from all series for three rotations, covering a period of 15 years, were 34.94, 35.35 and 38.30 per cent, respectively, for the first, second and third rotations. The writer is not inclined to attribute this to the residual effect of the fertilizers since in many cases the last crop on the treated pots was little larger than on the checks, indicating that there is little benefit from any residual nitrogen that

TABLE 12

Comparative availability of various nitrogenous fertilizers in soil under greenhouse conditions

Fifth crop (buckwheat)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
				Norfolk sand					
Unlimed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
1	Check	0.1			1.70		1.70		
2		1.0	0.55		1.67	1.68	16.70	9.20	
3	Ammonium phosphate	0.5			1.70		8.50		
4		0.4	0.45	−0.10	1.70	1.70	6.80	7.65	− 1.55
5	Ammonium sulfate	0.1			1.70		1.70		
6		0.1	0.10	−0.45	1.70	1.70	1.70	1.70	− 7.50
7	Dried blood	1.0			1.78		17.00		
8		0.4	0.70	0.15	1.70	1.74	6.80	11.90	2.70
9	Cottonseed meal	0.5			1.70		8.50		
10		0.5	0.50	−0.05	1.70	1.70	8.50	8.50	− 0.65
Limed									
11	Check	0.0							
12		0.0	0.0						
13	Ammonium phosphate	0.5			1.70		8.50		
14		0.5	0.50	0.50	1.70	1.70	8.50	8.50	8.50
15	Ammonium sulfate	0.2			1.70		3.40		
16		0.0	0.10	0.10		1.70		1.70	1.70
17	Dried blood	0.3			1.70		5.10		
18		0.5	0.40	0.40	1.70	1.70	8.50	6.80	6.80
19	Cottonseed meal	0.3			1.70		5.10		
20		0.5	0.40	0.40	1.70	1.70	8.50	6.80	6.80
				Sassafras loam					
Unlimed									
21	Check	2.9			1.68		48.72		
22		2.7	2.80		1.74	1.71	46.98	47.85	
23	Ammonium phosphate	6.1			1.76		107.36		
24		3.5	4.80	2.00	2.11	1.93	73.85	90.60	42.75
25	Ammonium sulfate	2.5			2.32		58.00		
26		4.5	3.50	0.70	2.46	2.39	110.70	84.35	36.50
27	Dried blood	3.2			1.68		53.76		
28		1.8	2.50	−0.30	2.26	1.97	40.68	47.22	− 0.63
29	Cottonseed meal	3.3			2.06		67.98		
30		4.4	3.85	1.05	1.74	1.90	76.56	72.27	24.42

TABLE 12—(Continued)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
				Sassafras loam					
Limed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
31	Check	1.9			1.75		33.25		
32		1.7	1.80		1.60	1.67	27.20	30.22	
33	Ammonium phosphate	lost							
34		lost							
35	Ammonium sulfate	lost							
36		2.2	2.20	0.40	1.44	1.44	31.68	31.68	1.46
37	Dried blood	lost							
38		3.4	3.40	1.60	1.42	1.42	48.28	48.28	18.06
39	Cottonseed meal	3.3			1.24		40.92		
40		2.6	2.95	1.15	1.55	1.39	40.30	40.61	10.39
				Norfolk sand					
Unlimed									
41	Check	1.2			1.80		21.60		
42		2.2	1.70		1.81	1.80	39.82	30.71	
43	Ammonium phosphate	0.6			1.70		10.20		
44		0.8	0.70	−1.00	1.70	1.70	13.60	11.90	−18.81
45	Ammonium sulfate	0.2			1.70		3.40		
46		0.0	0.10	−1.60	1.70	1.70	0.00	3.40	−27.31
47	Dried blood	0.2			1.70		3.40		
48		0.6	0.40	−1.30	1.70	1.70	10.20	6.80	−23.91
49	Cottonseed meal	0.4			1.70		6.80		
50		0.5	0.45	−1.25	1.70	1.70	8.50	7.65	−23.06
Limed									
51	Check	0.0	0.00						
52		0.0	0.00						
53	Ammonium phosphate	1.5							
54		lost	1.50	1.50	1.78	1.78	26.70	26.70	26.70
55	Ammonium sulfate	0.4			1.70		6.80		
56		0.3	0.35	0.35	1.70	1.70	5.10	5.95	5.95
57	Dried blood	0.6			1.70		10.20		
58		0.5	0.55	0.55	1.70	1.70	8.50	9.35	9.35
59	Cottonseed meal	0.2			1.70		3.40		
60		0.1	0.15	0.15	1.70	1.70	1.70	2.55	2.55

TABLE 12—(Concluded)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK
				Sassafras loam					
Unlimed									
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*
51	Check	1.0			1.86		18.60		
52		2.4	1.70		1.86	1.86	44.64	31.62	
63	Ammonium phosphate	6.9			2.44		168.36		
64		4.9	5.90	4.20	2.38	2.41	116.62	142.49	110.87
65	Ammonium sulfate	6.5			2.38		154.70		
66		3.5	5.00	3.30	2.52	2.45	88.20	121.45	89.83
67	Dried blood	4.5			2.25		101.25		
68		4.9	4.70	3.00	3.06	2.65	149.94	125.59	93.97
69	Cottonseed meal	5.4			1.54		83.16		
70		4.0	4.70	3.00	2.56	2.05	102.40	92.78	61.16
Limed									
71	Check	2.7			1.23		33.21		
72		2.5	2.60		1.33	1.28	33.25	33.23	
73	Ammonium phosphate	lost							
74		3.0	3.00	0.40	1.66	1.66	49.80	49.80	16.57
75	Ammonium sulfate	3.4			1.39		47.26		
76		3.0	3.20	0.60	1.73	1.56	51.90	49.58	16.35
77	Dried blood	3.9			1.21		47.19		
78		3.9	3.90	1.30	1.44	1.32	56.16	51.67	18.44
79	Cottonseed meal	3.6			1.55		55.80		
80		3.0	3.30	0.70	1.61	1.58	48.30	52.05	18.82

may be present. A better explanation for the higher recoveries of nitrogen for the second application of fertilizers lies in the depletion of available plant-food. A soil which has plenty of available nitrogen or at least a fairly large amount will not give high recoveries of nitrogen because the crops can get what they need from the soil organic matter. On the other hand, a crop grown on a poor soil will utilize the nitrogen applied very readily because there is no other sufficient supply available.

The summarized results in table 13 for the unlimed Norfolk sand with the lower rate of application show a lower availability for ammonium phosphate than ammonium sulfate, judging from the recovery of nitrogen. This is contrary to the results obtained for the first application of fertilizer. However, the crop yields are higher for ammonium phosphate, again illustrating the point previously brought out that frequently ammonium sulfate produces a smaller crop but with a higher per cent of nitrogen than does ammonium

TABLE 13

Comparative availability of various nitrogenous fertilizers in soil under greenhouse conditions. Summary—Fourth and fifth crops

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	TOTAL NITROGEN RECOVERED	AVERAGE	INCREASE OVER CHECK	PER CENT RECOVERED
	Norfolk sand (167 mgm. of nitrogen added)							
Unlimed								
		grams	*grams*	*grams*	*mgm.*	*mgm.*	*mgm.*	
1	Check	3.6			51.05			
2		2.5	3.05		40.19	45.57		
3	Ammonium phosphate	16.7			178.60			
4		16.1	16.40	13.35	160.66	169.63	124.06	74.28
5	Ammonium sulfate	13.8			157.88			
6		15.5	14.65	11.60	211.14	184.51	138.94	83.19
7	Dried blood	10.0			133.10			
8		8.8	9.40	6.35	108.44	120.77	75.20	45.03
9	Cottonseed meal	11.0			122.95			
10		11.0	11.0	7.95	105.10	114.02	68.45	40.99
Limed								
11	Check	5.0			67.50			
12		4.6	4.80		57.04	62.27		
13	Ammonium phosphate	18.6			193.12			
14		17.2	17.90	13.10	197.21	195.16	132.89	79.57
15	Ammonium sulfate	16.2			174.60			
16		16.7	16.45	11.65	178.69	176.64	114.37	68.48
17	Dried blood	12.0			126.78			
18		14.4	13.20	8.40	157.23	141.50	79.23	47.44
19	Cottonseed meal	14.0			140.73			
20		11.7	12.85	8.05	137.30	138.51	76.24	45.65
	Sassafras loam (167 mgm. of nitrogen added)							
Unlimed								
21	Check	4.0			73.58			
22		3.9	3.95		71.58	72.58		
23	Ammonium phosphate	11.5			227.78			
24		7.5	9.50	5.55	167.85	197.81	125.23	75.00
25	Ammonium sulfate	6.6			164.60			
26		6.7	6.65	2.70	166.14	165.37	92.79	55.56
27	Dried blood	5.8			123.70			
28		6.4	6.10	2.15	147.86	135.78	63.20	37.84
29	Cottonseed meal	6.5			141.26			
30		7.4	6.95	3.00	148.86	145.06	72.48	43.40

TABLE 13—(Continued)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	TOTAL NITROGEN RECOVERED	AVERAGE	INCREASE OVER CHECK	PER CENT RECOVERED
Limed								
		grams	*grams*	*grams*	*mgm.*	*mgm.*	*mgm.*	
31	Check	9.9			129.25			
32		10.4	10.15		124.64	126.94		
33	Ammonium phosphate	lost						
34								
35	Ammonium sulfate	20.2	20.20	10.05	254.88	254.88	127.94	76.61
36								
37	Dried blood	17.1	17.10	6.95	220.90	220.90	93.96	56.23
38								
39	Cottonseed meal	18.5			224.84			
40		19.1	18.80	8.65	230.05	227.44	100.50	60.18
	Norfolk sand (334 mgm. of nitrogen added)							
Unlimed								
41	Check	2.0			35.92			
42		2.2	2.10		39.82	37.87		
43	Ammonium phosphate	23.7			275.85			
44		23.5	23.60	21.50	286.00	280.92	243.05	72.77
45	Ammonium sulfate	14.4			264.68			
46		15.2	14.80	12.70	253.84	259.26	221.29	66.25
47	Dried blood	12.0			159.16			
48		13.5	12.75	10.65	175.79	167.47	129.60	38.80
49	Cottonseed meal	17.2			206.72			
50		15.3	16.25	14.15	202.38	204.55	166.68	49.90
Limed								
51	Check	3.3			43.23			
52		4.5	3.90		61.65	52.44		
53	Ammonium phosphate	31.0			360.05			
54		lost	31.00	27.10		360.05	307.61	92.09
55	Ammonium sulfate	24.4			297.20			
56		25.6	25.00	21.10	296.05	296.62	244.18	73.11
57	Dried blood	18.2			249.56			
58		20.7	19.45	15.55	263.02	256.29	203.85	61.03
59	Cottonseed meal	19.8			238.60			
60		22.1	20.95	17.05	248.10	243.35	190.91	57.16

TABLE 13—(Concluded)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	TOTAL NITROGEN RECOVERED	AVERAGE	INCREASE OVER CHECK	PER CENT RECOVERED
	Sassafras loam (334 mgm. of nitrogen added)							
Unlimed								
		grams	*grams*	*grams*	*mgm.*	*mgm.*	*mgm.*	
61	Check	3.1			58.71			
62		3.0	3.05		56.70	57.70		
63	Ammonium phosphate	12.9			311.76			
64		10.5	11.70	8.65	263.34	287.55	229.85	68.81
65	Ammonium sulfate	10.7			261.80			
66		8.9	9.70	6.65	230.68	246.24	188.54	56.45
67	Dried blood	6.9			163.69			
68		7.8	7.35	4.30	245.35	204.02	146.32	43.81
69	Cottonseed meal	10.9			221.21			
70		9.6	10.25	7.20	237.36	229.28	171.58	51.37
Limed								
71	Check	10.2			126.21			
72		11.7	10.95		157.47	141.84		
73	Ammonium phosphate	lost						
74		26.5	26.50	15.55	348.25	348.25	206.41	61.80
75	Ammonium sulfate	30.1			382.42			
76		34.0	32.05	21.10	386.70	384.56	242.72	72.67
77	Dried blood	26.9			357.69			
78		26.3	26.60	15.65	340.64	349.16	207.32	62.07
79	Cottonseed meal	24.2			278.28			
80		26.9	25.55	14.60	339.88	309.08	167.24	50.07

phosphate. Dried blood and cottonseed meal were decidedly lower in availability than either of the ammonium fertilizers.

At the higher rate of application of nitrogen in the absence of lime, ammonium phosphate gave nearly double the crop yield that ammonium sulfate did, but only about 6.5 per cent higher recovery of nitrogen. The high per cent of nitrogen in the crop from the ammonium sulfate pots nearly balanced the smaller crop yield. Cottonseed meal gave somewhat better results than dried blood, but both these organic fertilizers were decidedly lower in availability than the chemical fertilizers.

Where lime was applied to the Norfolk sand the crop yield and also the total recovery of nitrogen were always higher with ammonium phosphate than with ammonium sulfate. In this case the percentages of nitrogen in the crops grown from the two fertilizers were practically the same, contrary to the results on the unlimed soils. As shown in a later table ammonium sulfate increases the acidity of the soil to a slightly greater extent than does ammonium

phosphate. This may partially explain the results, but doubtless the differences noted may be attributed largely to the action of the sulfate and phosphate radicals.

The results from the unlimed Sassafras loam with both rates of application are consistent in the sense that the same relative results were obtained with both the high and the low rates of application. In the percentage recovery of nitrogen, ammonium phosphate was first, ammonium sulfate second, cottonseed meal third and dried blood fourth. With one exception the increase in crop yields over the checks was in the same order. With the higher rate of application cottonseed meal gave a slightly larger yield than ammonium sulfate but the difference is so slight that it may be neglected.

So many results were lost at the lower rate of application in the case of the limed Sassafras loam that very little can be said about it. Doubtless the yields would have been comparable with those obtained where the larger amounts of nitrogen were used. As usual, dried blood and cottonseed meal were not as efficient as ammonium sulfate but the difference was not as great here as in the previous results discussed. Both ammonium phosphate results were lost, one through the mistake in the application of the fertilizer and the other as a result of the "damping off" fungi. With the higher rate of application ammonium sulfate gave a larger crop yield and a higher recovery of nitrogen than ammonium phosphate. Dried blood showed unexpectedly good results, giving the same yield as ammonium phosphate and practically the same recovery of nitrogen. Cottonseed meal was somewhat less available than either of the other fertilizers used.

After the removal of the fifth crop from the pots the original fertilizer applications were repeated except that no lime was added. Corn was planted and harvested when about 18 inches high. The dry weights and percentages of nitrogen in the crop are given in table 14.

On the Norfolk sand, whether limed or unlimed, there was no great difference between the results from ammonium sulfate and from ammonium phosphate. Dried blood was usually not quite as efficient as cottonseed meal but both were inferior to the two ammonium salts.

On the Sassafras loam the two organic fertilizers gave larger yields than the chemical fertilizers on the unlimed soil, but were usually not as efficient in the presence of lime. Ammonium sulfate was somewhat more effective than ammonium phosphate, which is partially in contradiction to the previous crops. Possibly the residual effect, if there is one, together with the fact that a different kind of crop was grown, accounts for the difference.

In table 15 a summary is given of the results for the six crops grown on the same soils. In calculating the relative increases in yield of dry matter and the relative recoveries of nitrogen, the results for the two different rates of application are averaged. Ammonium phosphate is then given a value of 100 and the other fertilizers figured on this basis.

In figures 6 and 7 the relative increases in dry matter over the checks and

TABLE 14

Comparative availability of various nitrogenous fertilizers in soil under greenhouse conditions. Sixth crop (corn)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK	PER CENT RECOVERED
	Norfolk sand (167 mgm. of nitrogen added)									
Unlimed										
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*	
1	Check	4.3			1.13		48.59			
2		4.2	4.25		1.16	1.15	48.72	48.65		
3	Ammonium phosphate	9.0			1.22	1.24	109.80			
4		10.1	9.55	5.30	1.25	1.24	126.25	118.02	69.37	41.54
5	Ammonium sulfate	9.0			1.43		128.70			
6		9.9	9.45	5.20	1.29	1.36	127.71	128.20	79.55	47.63
7	Dried blood	6.5			1.39		90.35			
8		6.6	6.55	2.30	1.53	1.46	100.98	95.66	47.01	28.15
9	Cottonseed meal	6.5			1.39		90.35			
10		8.5	7.50	3.25	1.30	1.35	110.5	100.42	51.77	31.00
Limed										
11	Check	4.1			1.27		52.07			
12		3.6	3.85		1.37	1.32	49.32	50.69		
13	Ammonium phosphate	11.0	10.00	6.15	1.32	1.39	145.20	138.30	87.61	52.46
14		9.0			1.46		131.40			
15	Ammonium sulfate	7.7	9.05	5.20	1.41	1.40	108.57	126.56	75.87	45.43
16		10.4			1.39		144.56			
17	Dried blood	7.0	7.00	3.15	1.51	1.49	105.70	103.95	53.26	31.89
18		7.0			1.46		102.20			
19	Cottonseed meal	8.0	8.45	4.60	1.37	1.34	109.60	112.65	61.96	37.10
20		8.9			1.30		115.70			
	Sassafras loam (167 mgm. of nitrogen added)									
Unlimed										
21	Check	3.2			1.72		55.04			
22		3.0	3.10		1.78	1.75	53.40	54.22		
23	Ammonium phosphate	4.6			2.03		93.38			
24		4.2	4.40	1.30	1.93	1.98	81.06	87.22	33.00	19.76
25	Ammonium sulfate	5.5			2.06		113.30			
26		4.9	5.20	2.10	1.92	1.99	94.08	103.69	49.47	29.60
27	Dried blood	5.3			1.85		98.05			
28		6.0	5.65	2.55	1.82	1.84	109.2	103.62	49.40	29.51
29	Cottonseed meal	5.5			1.88		103.40			
30		5.3	5.40	2.30	1.75	1.82	92.75	98.07	43.85	26.26

TABLE 14—(Continued)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK	PER CENT RECOVERED
	Sassafras loam (167 mgm. of nitrogen added)									
Limed										
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*	
31	Check	7.0			1.30		91.00			
32		6.4	6.70		1.32	1.31	84.48	87.74		
33	Ammonium phosphate	lost								
34		lost								
35	Ammonium sulfate	lost								
36		12.00	12.00	5.30	1.44	1.44	172.80	172.80	85.06	50.93
37	Dried blood	lost								
38		11.1	11.10	4.40	1.36	1.36	150.96	150.96	63.22	37.85
39	Cottonseed meal	9.0			1.57		141.30			
40		12.0	10.50	3.80	1.43	1.50	171.60	156.45	68.71	41.14
	Norfolk sand (334 mgm. of nitrogen added)									
Unlimed										
41	Check	5.4			1.34		72.36			
42		4.5	4.95		1.30	1.32	58.50	65.43		
43	Ammonium phosphate	16.7			1.53		255.51			
44		15.1	15.90	10.95	1.60	1.57	241.60	248.55	183.12	54.82
45	Ammonium sulfate	14.6			1.78		259.88			
46		18.5	16.55	11.60	1.41	1 60	256.85	258.36	192.93	57.76
47	Dried blood	13.5			1.53		206.55			
48		11.0	12.25	7.30	1.43	1.48	157.30	181.92	116.49	34.87
49	Cottonseed meal	15.0			1.36		204.00			
50		16.5	15.75	10.80	1:32	1.34	217.80	210.90	145.47	43.55
Limed										
51	Check	5.0			1.48		74.00			
52		5.8	5.40		1.26	1.37	73.08	73.54		
53	Ammonium phosphate	17.6	17.60	12.20	1.51	1.51	265.76	265.76	192.22	57.55
54		lost								
55	Ammonium sulfate	17.7			1.47		260.19			
56		18.7	18.20	12.80	1.47	1.47	274.89	267.54	194.00	58.08
57	Dried blood	13.3			1.72		228.76			
58		16.9	15.10	9.70	1.32	1.52	223.08	225.92	152.38	45.62
59	Cottonseed meal	13.7			1.58		216.46			
60		13.3	13.50	8.10	1.41	1.50	187.53	201.99	128.45	38.46

TABLE 14—(Concluded)

Sassafras loam (334 mgm. of nitrogen added)

POT NO.	TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK	NITROGEN	AVERAGE	TOTAL NITROGEN	AVERAGE	INCREASE OVER CHECK	PER CENT RECOVERED
Unlimed										
		grams	*grams*	*grams*	*per cent*	*per cent*	*mgm.*	*mgm.*	*mgm.*	
61	Check	4.0			1.43		57.20			
62		4.4	4.20		1.43	1.43	62.92	60.06		
63	Ammonium phosphate	5.1			2.16		110.16			
64		7.3	6.20	2.00	2.04	2.10	148.92	129.54	69.48	20.80
65	Ammonium sulfate	7.2			1.81		130.32			
66		6.9	7.05	2.85	1.85	1.83	127.65	128.98	68.92	20.63
67	Dried blood	9.7	9.70	5.50	2.03	2.03	196.91	196.91	136.85	40.97
68		lost								
69	Cottonseed meal	8.6			1.90		163.40			
70		7.5	8.05	3.85	2.09	2.00	156.75	160.07	100.01	29.94
Limed										
71	Check	5.2			1.26		65.52			
72		5.1	5.15		1.30	1.28	66.30	65.91		
73	Ammonium phosphate	lost								
74		14.9	14.90	9.75	1.50	1.50	223.50	223.50	157.59	47.18
75	Ammonium sulfate	18.5			1.43		264.55			
76		18.0	18.25	13.10	1.46	1.45	262.80	263.67	197.76	59.21
77	Dried blood	14.5			1.64		237.80			
78		16.1	15.30	10.15	1.57	1.61	252.77	245.28	179.37	53.70
79	Cottonseed meal	12.7			1.58		200.66			
80		13.6	13.15	8.00	1.62	1.60	220.32	210.49	144.58	43.29

the relative recoveries of nitrogen with the four fertilizers are shown diagrammatically for both the Norfolk and Sassafras loam soils. In this case the results for the two different rates of application are shown separately.

Referring to table 15 it will be noted that ammonium phosphate gave appreciably larger increases in dry matter on both the limed and unlimed Norfolk soil than did ammonium sulfate, and very much greater yields than the two organic fertilizers. On the Sassafras loam the crop yields are quite peculiar, ammonium phosphate giving more than twice the increases of ammonium sulfate in the absence of lime, while where lime is present there is a 32 per cent increase in favor of ammonium sulfate.

The relative yields of dry matter are quite interesting when compared with the recoveries of nitrogen. In the presence of lime the fertilizers which gave the greatest increases in crop yields also gave the highest recoveries of nitrogen, as might be expected, but when lime was absent the figures were somewhat different, especially on the Sassafras loam. With ammonium phosphate

TABLE 15

Comparative availability of various nitrogenous fertilizers in soil under greenhouse conditions. Summary—Six crops

Increase in dry weights

Norfolk sand

MGM. OF NITROGEN APPLIED	UNLIMED				LIMED			
	Ammonium phosphate	Ammonium sulfate	Dried blood	Cottonseed meal	Ammonium phosphate	Ammonium sulfate	Dried blood	Cottonseed meal
501 .... (gm)	31.25	28.10	16.06	20.52	33.90	28.67	22.41	18.46
1002 .... (gm)	62.92	52.81	38.56	41.45	65.41	58.77	44.71	38.50
Total ... (gm)	94.17	80.91	54.62	61.97	99.31	87.44	67.12	56.96
Ratio	100.00	85.90	58.00	65.80	100.00	88.00	67.60	57.30

Sassafras loam

501 .... (gm)	12.41	5.46	10.08	9.59		17.51	14.92	11.52*
1002 .... (gm)	28.03	13.35	20.14	20.36	33.23	43.99	29.79	26.58
Total ... (gm)	40.44	18.81	30.22	29.95				
Ratio	100.00	46.50	74.70	74.60	100.00	132.40	89.60	80.00

Percentage of nitrogen recovered

Norfolk sand

MGM. OF NITROGEN APPLIED	UNLIMED				LIMED			
	Ammonium phosphate	Ammonium sulfate	Dried blood	Cottonseed meal	Ammonium phosphate	Ammonium sulfate	Dried blood	Cottonseed meal
	per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
501.....	60.77	62.41	41.30	38.22	65.41	55.12	42.87	36.21
1002.....	64.15	62.24	40.76	45.74	73.19	64.64	50.05	44.61
Average nitrogen recovery	63.02	62.30	40.94	43.23	70.60	61.47	47.66	41.81
Relative availability	100.00	98.90	64.90	68.60	100.00	87.10	67.50	59.20

Sassafras loam

501.....	52.11	46.81	38.33	36.47		64.27	48.99	38.71*
1002.....	51.02	53.71	49.97	41.69	42.26	50.75	39.24	31.32
Average nitrogen recovery	51.38	51.41	46.09	39.95				
Relative availability.....	100.00	100.10	89.70	77.70	100.00	120.10	92.80	74.10

*Incomplete line not included in calculating ratios.

taken as 100, ammonium sulfate showed a relative crop yield of about 44 in one case and 47.6 in another. The yields from dried blood and cottonseed

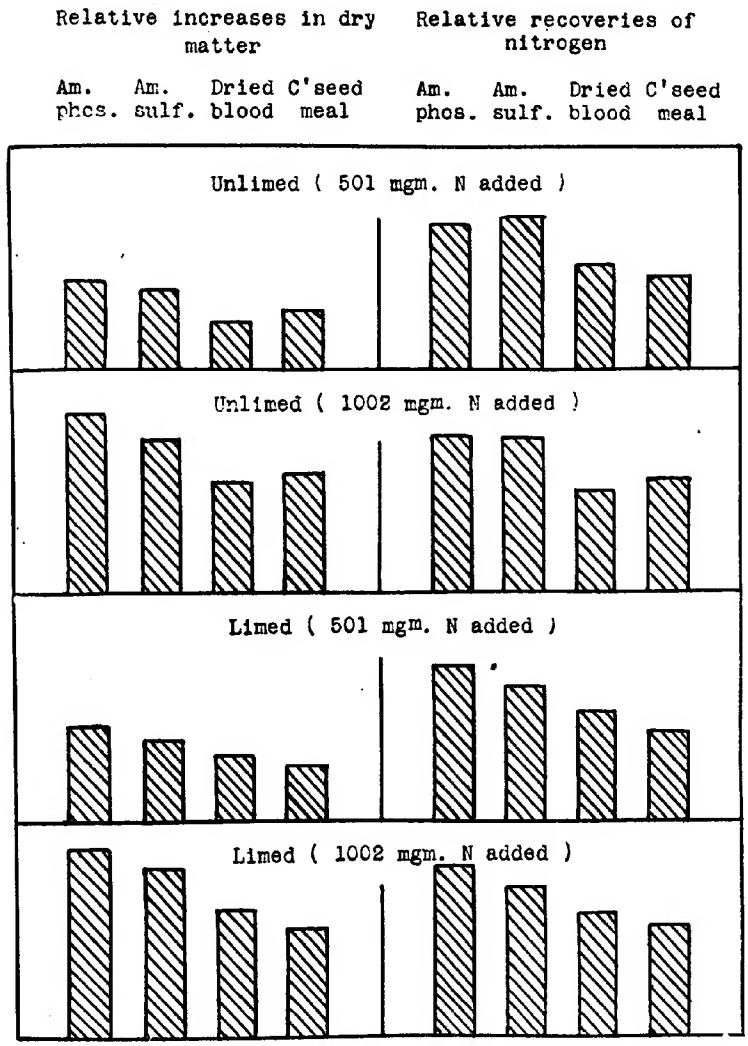

FIG. 6. DIAGRAM SHOWING THE AVAILABILITY OF VARIOUS NITROGENOUS FERTILIZERS IN NORFOLK SAND

meal also were greater than those from ammonium sulfate, but not equal to those from ammonium phosphate. Of course the increases over the checks were quite small on this soil as compared with those on the Norfolk sand, but

nevertheless they are great enough to bring out strikingly that under very acid conditions ammonium sulfate may show a comparatively small crop, but

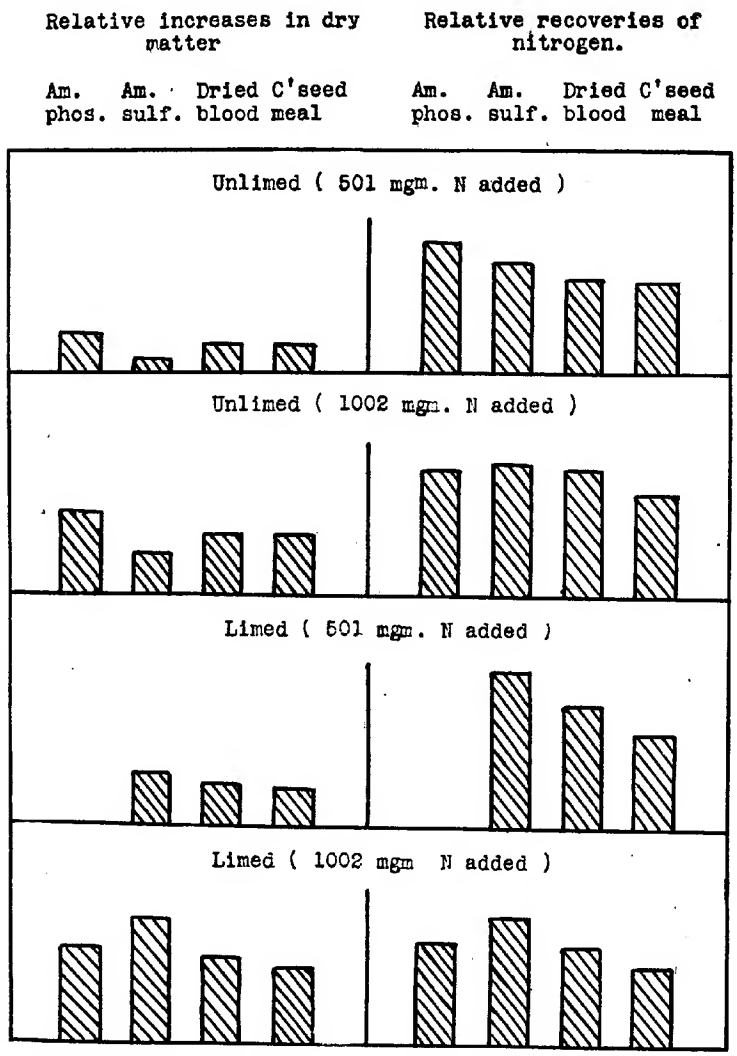

FIG. 7. DIAGRAM SHOWING THE AVAILABILITY OF VARIOUS NITROGENOUS FERTILIZERS IN SASSAFRAS LOAM

on analysis a higher percentage recovery of nitrogen than other fertilizers. It will be remembered that this soil contained a high initial per cent of nitrogen.

Many investigators have recorded the fact that the use of nitrogenous

fertilizers increases the content of nitrogen in the dry matter of plants and necessarily lowers the percentage of carbohydrates. Among these may be mentioned Lipman (21), Beseler and Maercher (1), Ritthausen and Pott (28) and Wagner (34). The latter investigator emphasized the ability of plants to produce large quantities of dry matter with a low nitrogen content when conditions are favorable or a small amount of dry matter with a high nitrogen content when there is an excess of available nitrogen present. The latter condition is quite common where a nitrogenous fertilizer is applied to a soil which is already rich in nitrogen. Adolph Mayer (24) found that while the application of readily-soluble nitrogen may increase the nitrogen content of the plant, there may not necessarily be an increase in protein nitrogen. In an experiment on the relation of lime to the availability of ammonium sulfate, Cook and Allison (4) showed that, while the use of lime in connection with ammonium sulfate may greatly increase the crop yield, the analysis of the crop does not always show a higher recovery of nitrogen in such cases. Similar results to those reported in this thesis were obtained in many cases. While the writers mentioned have shown that the use of the readily available fertilizers, usually sodium nitrate, may increase the per cent of nitrogen in the crop, the enormous increase reported here seems to be wholly out of proportion. In another experiment, the results of which have not yet been published, the writer was able to grow small buckwheat plants in an acid loam soil for a few weeks in the presence of an unusually high percentage of ammonium sulfate, which on analysis showed 16 per cent of nitrogen, while plants grown in a similar soil in the absence of nitrogenous fertilizers showed 0.8 per cent of nitrogen. Evidently the nitrogen represented by this 2000 per cent increase over the check was not present in the plant as protein nitrogen. Whether the same results would have been obtained with ammonium phosphate is, of course, only a matter of conjecture, but the results reported in this thesis indicate that the ammonium phosphate would have behaved in a more normal manner.

As an average of all the conditions, the recoveries of the nitrogen applied to the Norfolk sand were 65.88, 61.10, 43.74 and 41.19, respectively, for ammonium phosphate, ammonium sulfate, dried blood and cottonseed meal. The highest recovery in any case was 73.19 per cent on the limed soil. On the Sassafras loam the average recoveries were 48.46, 50.42, 42.51, and 36.49 per cent for ammonium phosphate, ammonium sulfate, dried blood and cottonseed meal, respectively, the highest being 53.71 per cent on an unlimed soil. In calculating these averages the incomplete set of figures for the smaller application of nitrogen on the limed soil are omitted.

The question naturally arises, what has become of the remainder of the nitrogen applied? No analyses of the soils from the various pots were made to determine if there was more or less nitrogen at the end of the experiment than at the beginning, but this was scarcely necessary for the mere fact that plants would not grow in the pots after removing five crops is in itself sufficient

proof that the supply of nitrogen had decreased very markedly. Even if it were present in an unavailable form it would have soon been made available. Under greenhouse conditions ammonification and nitrification can proceed at a very rapid rate. In explaining the fact that under the most favorable conditions usually not more than 60 per cent of the nitrogen added is recovered in the crop the usual causes set forth have been leaching, fixation in the soil, loss as ammonia, and loss as nitrogen gas. The last two have not been emphasized to any great extent because leaching and fixation by the zeolites and as proteins by the soil microörganisms seemed more plausible.

In this experiment leaching cannot be considered since the plants were grown in pots having no provision for drainage. The second explanation, namely, fixation, does not seem to be of importance, considering that five crops were grown and any nitrogen fixed in the soil, especially as protein, would probably have become available for the plants during the course of the experiment. The most logical conclusion, then, is that nitrogen escaped as free nitrogen gas or as ammonia.

Among the many investigators who have made a study of the loss of ammonia from soils, the following may be mentioned. Hals (7) found that ammonia is lost from loamy soils rich in lime when small quantities of ammonium sulfate are added and the soil is exposed to wind and sun. Wagner (35) attributes the lower efficiency of ammonium sulfate to be due to the escape of ammonia into the air. Kretschiner (13) states that ammonium sulfate should not be used as a top dressing with soils rich in lime. Schäfer-Heide (30) observed that a high lime content and high temperature favored loss of ammonia, especially on light soils. Schreiber (31) conducted pot experiments with oats to determine the effect of calcium carbonate on ammonium sulfate and found that where ammonium sulfate was used as a top-dressing on soils rich in lime decided losses of ammonia occurred. Lemmermann (14) states that the danger of loss of ammonia is small provided the ammonium sulfate is thoroughly incorporated with the soil and the application does not follow liming too closely. Ehrenberg (5) states that loss of ammonia by volatilization is to be feared only in the case of sandy soils which are rich in calcium carbonate, contain little humus and are poor in zeolitic compounds. Even in such soils the loss is not appreciable except at high temperatures and in dry soils containing high contents of ammonia. Lemmerman (15) found that the addition of calcium carbonate to soil in pots reduces the volatilization of ammonium carbonate and increases the absorptive power of the soil for ammonia. Caustic lime exerted an opposite effect.

Numerous studies on the loss of nitrogen gas from soils have been made but only a few references can be given in this discussion. The experiments of Immendorff (10) are significant in that they point to a loss of free nitrogen during the nitrification of ammonium salts. Wilfarth, Römer and Wimmer (36, p. 26) point out that there may be a volatilization of elementary nitrogen during respiration. Lipman and Blair (22) in a discussion of various phases

of the nitrogen problem, state that "there seems to be more or less justification for the assumption made by these investigators (Wilfarth, Römer and Wimmer) for it is quite possible that in the respiration processes there may be destruction, not only of the non-nitrogenous organic compounds, but also of nitrogenous organic compounds, like amino acids or their derivatives. At any rate it has been known for years (2) that there is a loss of nitrogen in the germination of seeds, and the analogy might well be extended to the plant after germination." The same investigators, commenting upon the lower availability of dried blood nitrogen as compared with ammonium sulfate nitrogen state that in the process of decay there are many stages where loss of elementary nitrogen, is liable to occur and the greater the resistance to decay the greater is the danger of loss. Also, in nitrification processes a portion of the nitrogen is set free. The loss with ammonium salts is slight but probably more appreciable with organic combinations.

From the data reported in this paper, obtained with the use of ammonium phosphate, ammonium sulfate, dried blood and cottonseed meal, and in the light of previous work done by various investigators, the writer is inclined to attribute the failure to obtain a complete recovery of the nitrogen applied to the soil in the crops largely to a loss as ammonia or nitrogen gas. Half of the pots received an application of calcium carbonate, yet the recovery of nitrogen from these pots was usually larger than from the unlimed pots; therefore the presence of the lime cannot have been the primary cause of loss of nitrogen. Possibly calcium oxide would have caused a greater loss than occurred on the untreated soils. It would seem that either there is an appreciable loss of ammonia from all soils, both sand and clay, even when kept moist and relatively free from air circulation, or else there is a loss as nitrogen gas from the soil by transpiration. While it has not yet been proved that nitrogen is evolved from plants, there is plenty of evidence strongly indicating that such is the case.

Effect of fertilizers on soil reaction

At the end of a little over one year from the time the experiment on the comparative availability of various nitrogenous fertilizers had been started, and directly following the harvesting of the fifth crop, the lime requirement of the soils in all of the pots was determined. The samples of soil for these analyses were obtained by removing the 20 pounds of soil from the pots, thoroughly mixing and air-drying. The Veitch method for the determination of acidity was used throughout the work. The data are given in table 16, the results being omitted for pots 33, 35 and 37 which, it will be remembered, accidently received an extra application of fertilizer. In other cases fungi injured the crop, but the lime requirements are given regardless of this fact.

In general, the results show that ammonium sulfate and ammonium phosphate increase the acidity to a greater extent than dried blood or cottonseed meal. Usually ammonium sulfate caused a greater acidity than ammonium

TABLE 16

Effect of fertilizers on soil reaction

POT NO.	TREATMENT	MGM. NITROGEN ADDED	MGM. NITROGEN RECOVERED	RESIDUE IN SOIL	LIME REQUIRED AT START. LBS. CaO PER ACRE	LIME REQUIRED AT END. LBS. CaO PER ACRE	INCREASE LBS. PER ACRE	AVERAGE INCREASE LBS. PER ACRE	AVERAGE INCREASE DUE TO FERTILIZERS. LBS. PER ACRE
				Norfolk sand					
					Acid	*Acid*	*Acid*		
1	Check	0	162.39		400	3300	3700		
2		0	142.70		400	3500	3900	3800	
3	Ammonium phosphate	334	394.62	91.92	400	3900	4300		
4		334	380.70	105.84	400	4100	4500	4400	600
5	Ammonium sulfate	334	360.74	125.80	400	4300	4700		
6		334	410.66	75.88	400	4300	4700	4700	900
7	Dried blood	334	314.26	172.28	400	3900	4300		
8		334	310.61	175.93	400	4000	4400	4350	550
9	Cottonseed meal	334	302.00	184.54	400	3300	3700		
10		334	282.80	203.74	400	3500	3900	3800	000
					Alkaline	*Acid*	*Acid*		
11	Check	0	256.47		2240	900	3140		
12		0	229.06		2240	1000	3240	3190	
13	Ammonium phosphate	334	484.06	92.70	2240	1000	3240		
14		334	482.01	94.75	2240	1200	3440	3340	150
15	Ammonium sulfate	334	446.88	129.88	2240	1300	3540		
16		334	439.23	137.53	2240	1300	3540	3540	350
17	Dried blood	334	393.16	183.60	2240	900	3140		
18		334	416.41	160.35	2240	1300	3540	3340	150
19	Cottonseed meal	334	366.73	210.03	2240	1500	3740		
20		334	358.73	218.02	2240	1500	3740	3740	550
				Sassafras loam					
					Acid	*Acid*			
21	Check	0	454.74		2800	6100	3300		
22		0	471.14		2800	6300	3500	3400	
23	Ammonium phosphate	334	721.86	75.08	2800	5900	3100		
24		334	627.06	169.88	2800	6500	3700	3400	000
25	Ammonium sulfate	334	623.31	173.63	2800	6200	3400		
26		334	672.79	124.15	2800	6500	3700	3550	150
27	Dried blood	334	561.90	235.04	2800	6500	3700		
28		334	649.45	147.49	2800	6300	3500	3600	200
29	Cottonseed meal	334	615.50	181.44	2800	6100	3300		
30		334	588.15	208.79	2800	6100	3300	3300	−100

TABLE 16—(Continued)

POT NO.	TREATMENT	MGM. NITROGEN ADDED	MGM. NITROGEN RECOVERED	RESIDUE IN SOIL	LIME REQUIRED AT START. LBS. CaO PER ACRE	LIME REQUIRED AT END. LBS CaO PER ACRE	INCREASE LBS. PER ACRE	AVERAGE INCREASE LBS. PER ACRE	AVERAGE INCREASE DUE TO FERTILIZERS LBS. PER ACRE
				Sassafras loam					
					Alkaline	*Acid*			
31	Check	0	599.25		2240	1800	4040		
32		0	606.06		2240	1600	3840	3940	
33	Ammonium phosphate	334	lost		2240	lost			
34		334	lost		2240	2300	4540	4540	600
35	Ammonium sulfate	334	lost		2240	lost			
36		334	838.99	97.26	2240	3300	5540	5540	1600
37	Dried blood	334	lost		2240	lost			
38		334	784.93	151.72	2240	1700	3940	3940	0000
39	Cottonseed meal	334	712.98	223.67	2240	1500	3740		
40		334	742.82	193.88	2240	1900	4140	3940	000
				Norfolk sand					
					Acid	*Acid*			
41	Check	0	164.80		400	3700	4100		
42		0	163.93		400	3900	4300	4200	
43	Ammonium phosphate	668	624.29	208.07	400	4800	5200		
44		668	624.34	208.02	400	4900	5300	5250	1050
45	Ammonium sulfate	668	597.27	235.09	400	5200	5600		
46		668	597.10	235.26	400	4800	5200	5400	1200
47	Dried blood	668	443.38	388.98	400	4400	4800		
48		668	469.21	363.15	400	4300	4700	4750	550
49	Cottonseed meal	668	471.52	360.84	400	3700	4100		
50		668	482.92	349.44	400	3700	4100	4100	−100
					Alkaline	*Acid*			
51	Check	0	216.88		2240	1300	3540		
52		0	220.06		2240	1300	3540	3540	
53	Ammonium phosphate	668	761.85	124.62	2240	1800	4040		
54		668	lost		2240	1700	3940	3990	450
55	Ammonium sulfate	668	670.96	215.51	2240	2100	4340		
56		668	673.45	213.02	2240	2200	4440	4390	850
57	Dried blood	668	569.69	316.78	2240	1700	3940		
58		668	565.63	320.84	2240	1700	3940	3940	400
59	Cottonseed meal	668	539.54	346.93	2240	1700	3940		
60		668	534.42	352.05	2240	1500	3740	3840	300

TABLE 16—(Concluded)

POT NO.	TREATMENT	MGM. NITROGEN ADDED	MGM. NITROGEN RECOVERED	RESIDUE IN SOIL	LIME REQUIRED AT START. LBS. CaO PER ACRE	LIME REQUIRED AT END. LBS. CaO PER ACRE	INCREASE LBS. PER ACRE	AVERAGE INCREASE LBS. PER ACRE	AVERAGE INCREASE DUE TO FERTILIZERS LBS. PER ACRE
				Sassafras loam					
					Acid	*Acid*			
61	Check	0	435.66		2280	6300	4020		
62		0	488.98		2280	6300	4020	4020	
63	Ammonium phosphate	668	923.11	207.21	2280	6700	4420		
64		668	885.18	245.14	2280	6900	4620	4520	500
65	Ammonium sulfate	668	932.60	197.72	2280	6400	4120		
66		668	930.65	199.67	2280	6600	4320	4220	200
67	Dried blood	668	784.86	345.46	2280	6400	4120		
68		668	lost		2280	6500	4220	417[illegible]	150
69	Cottonseed meal	668	756.03	374.29	2280	6400	4120		
70		668	804.17	326.15	2280	6600	4320	4220	200
					Alkaline	*Acid*			
71	Check	0	672.58		2240	1700	3940		
72		0	865.99		2240	1700	3940	3940	
					Alkaline	*Acid*			
73	Ammonium phosphate	668	lost		2240	2500	4740		
74		668	1035.45	402.83	2240	2600	4840	4790	850
75	Ammonium sulfate	668	1089.51	347.77	2240	2800	5040		
76		668	1070.52	366.76	2240	2800	5040	5040	1100
77	Dried blood	668	974.27	463.01	2240	1900	4140		
78		668	992.06	445.22	2240	1700	3940	4040	100
79	Cottonseed meal	668	893.36	543.92	2240	1700	3940		
80		668	983.72	453.56	2240	1800	4040	3990	50

phosphate. The two organic fertilizers showed very different behavior under different conditions. Sometimes dried blood showed a greater development of acid than cottonseed meal and again the reverse was true, or perhaps no acidity developed at all above that in the checks.

Averaging the results of all determinations we find that the increase in lime requirement over the checks due to ammonium sulfate was 793.7 pounds of CaO, with ammonium phosphate 525 pounds, with dried blood 262.5 pounds, and with cottonseed meal 112.5 pounds. These results are about what would be expected and are in agreement with other work as far as ammonium sulfate and the organic forms of nitrogen are concerned. As far as the writer knows no data have been reported for ammonium phosphate. It is impossible in this paper to take up a discussion of the acidity work which has been carried out with the other fertilizers.

Comparative availability of the nitrogen in sodium nitrate, "Ammo-Phos," water-soluble "Ammo-Phos," and water-insoluble "Ammo-Phos" in sand cultures

Ten-pound portions of very poor red sand were weighed out into pots and the various fertilizers added, including sodium nitrate, "Ammo-Phos," and water-soluble and water insoluble "Ammo-Phos." Enough of each of these was applied so that each treated pot contained 100 mgm. of nitrogen. All pots received 1 gm. of KCl and 10 gm. of calcium carbonate together with enough acid phosphate to make the total phosphorus equal to that in water-soluble "Ammo-Phos." The dry weights of the buckwheat are given in table 17.

TABLE 17

Availability of different portions of "Ammo-Phos"

TREATMENT	YIELD	AVERAGE	INCREASE OVER CHECK
	grams	*grams*	*grams*
Check	4.0 3.1	3.55	
"Ammo-Phos"	14.9 lost	14.90	11.35
Water-soluble "Ammo-Phos"	14.5 14.0	14.25	10.70
Water-insoluble "Ammo-Phos"	lost 13.8	13.80	10.25
Sodium nitrate	15.0 13.5	14.25	10.70

From table 17 it will be noticed that there was practically no difference in the availability of the various forms of nitrogen. For some unknown reason "Ammo-Phos" was slightly better than sodium nitrate and even better than the water-insoluble portion of "Ammo-Phos." The water insoluble "Ammo-Phos" was nearly as good as the other forms of nitrogen, the difference being practically within experimental error.

General review and discussion of nitrogen availability experiments

The pot experiments have shown rather definitely the availability of the ammonium phosphate fertilizer as compared with other common nitrogenous fertilizers. It has been brought out that in general the nitrogen in ammonium phosphate gives at least as good results as that in ammonium sulfate and often slightly higher yields under greenhouse conditions. In the one experiment in sand in the greenhouse, sodium nitrate produced about the same increase in yield as ammonium phosphate. It is to be regretted that sodium

nitrate was not also included in the greenhouse soils experiment, but by comparing the results obtained when ammonium sulfate and the organic fertilizers were used, with the extensive experiments of other investigators, we gain a fair idea of the relative value of the nitrogen in ammonium phosphate as compared with that in the other common fertilizers. Since ammonium phosphate gives at least as good results as ammonium sulfate, then by considering the latter in relation to sodium nitrate we can gain a fair idea of what could be expected if ammonium phosphate were directly compared with sodium nitrate.

While it might be desirable to include a bibliography of the question of availability at this point, it is impossible to do so for want of space. However, the writer has reviewed the literature rather thoroughly and as a general summary it may be stated that nitrate nitrogen usually gives the best results, ammonia nitrogen ranks second, following by calcium cyanamid and the various organic fertilizers. The data reported in this paper are in general agreement with the average results of other investigators although, of course, ammonium phosphate was not included in their availability experiments.

The work of the various research men has shown better results for sodium nitrate and ammonium sulfate than for the organic form of nitrogen, but there is wide disagreement when we come to contrast ammonium sulfate and sodium nitrate. Some crops were found to prefer ammonia and others nitrate, but the soil and weather conditions are just as important as the crop or more so. Almost invariably the value of ammonium sulfate was relatively higher in wet seasons, while sodium nitrate was more or less independent of moisture, within certain limits, of course. Lime also favors ammonia nitrogen because it allows nitrification to proceed more rapidly through its neutralization of the acids present. Usually early spring applications were more effective than later applications. Several writers have cautioned against the use of large amounts of ammonium salts.

From the standpoint of ammonium phosphate any availability experiments which include ammonium sulfate are important since we can reasonably predict that the phosphate salt will behave in a similar manner to ammonium sulfate and acid phosphate. The data of this thesis for the most part substantiate this assumption. Perhaps the salt will give slightly better results than ammonium sulfate because there is no sulfate radical left in the soil to increase the acidity.

Comparative availability of various phosphate fertilizers in soil under greenhouse conditions

The procedure followed for this experiment was practically a duplication of that used in the nitrogen availability experiment reported in tables 7 to 15, except that phosphorus was the limiting factor instead of nitrogen. The same soils, namely, Norfolk sand and Sassafras loam were chosen. The pots

held only 10 pounds of soil. The fertilizers applied in addition to ammonium phosphate were acid phosphate (17.82 per cent P_2O_5), basic slag (19.04 per cent P_2O_5) and Tennessee raw rock phosphate (31.41 per cent P_2O_5). The rates of application were 0.5 gm. of acid phosphate to one-half of the pots and 1.0 gm. to the remainder, equivalent amounts of phosphorus being added for all treatments. Each pot received 1.3 gm. of potassium chloride. Three grams of ammonium sulfate also were added to all except the ammonium phosphate pots, in which case allowance was made for the nitrogen in the fertilizer. Half of the pots were limed and half left unlimed, as was done in the previous availability experiment. Three crops of buckwheat were grown in all of the pots but only one application of fertilizer was made. The dry weights of the plants are given in table 18 together with the average results for the three crops.

It will be noticed from the table that a few of the duplicate determinations do not agree as closely as might be desired. This is largely explained by the fact that the seed on these pots was not as good as it should have been. A few pots did not have as many plants as the others and consequently did not show quite as large a yield as would otherwise have been the case. However, it will be observed that usually when the yield of the first crop was low the yields for the next two crops tend to even up the differences and the final averages show reasonably close agreement for the duplicate pots. On the unlimed Sassafras loam the agreement between the duplicate treatments was not always close but this was due to the high acidity of the soil. When the fertilizers were added the increased amount of soluble salts applied only served to increase further the injury of the acids already present. Some of the plants on these pots were dead when harvested. None of the increases over the checks were very large, and for this reason it is impossible to draw any sharp distinctions in the availability of the phosphorus of the different fertilizers. The soils themselves contained practically all of that element needed for ordinary crop growth.

The unlimed Norfolk sand at the lower rate of application showed practically no difference in the availability of the phosphorus in ammonium phosphate, acid phosphate and basic slag. The yield from raw rock phosphate was slightly below the others. At the higher rate of application on this soil the yields were less than with the smaller amount of phosphorus present. Doubtless the higher concentration of salts in these pots was injurious.

Where lime was added to the Norfolk sand the yields were much larger than in the absence of lime, but the effect of the various phosphorus-containing fertilizers is negligible in this case. They probably increased the availability of the phosphorus in the soil. In some cases the addition of the fertilizers actually decreased the yield slightly, but the differences are within experimental error.

On the unlimed Sassafras loam at the lower rate of application acid phosphate showed the highest availability, ammonium phosphate second, and the

TABLE 18

Availability of phosphate fertilizers under greenhouse conditions

TREATMENT	FIRST CROP		SECOND CROP		THIRD CROP		AVERAGE		
	Weight	Average	Weight	Average	Weight	Average	Weight	Average	Increase over check
Norfolk sand (86.1 mgm. of P_2O_5 added)									
Unlimed									
	grams	*grams*	*grams*	*grams*	*grams*	*grams*	*grams*	*grams*	*grams*
Check	23.6 22.2	22.90	8.5 3.1	5.80	1.3 1.2	1.25	33.4 26.5	29.95	
Ammonium phosphate	30.0 26.4	28.20	8.8 7.4	8.10	2.5 1.6	2.05	41.3 35.4	38.35	8.40
Acid phosphate	25.0 25.5	25.25	10.0 14.0	12.00	1.2 1.4	1.30	36.2 40.9	38.55	8.60
Basic slag	26.0 20.4	23.20	11.5 14.2	12.85	1.5 1.4	1.45	39.0 35.6	37.30	7.35
Raw rock phosphate	21.5 27.6	24.55	12.3 5.8	9.05	1.4 2.5	1.95	35.2 35.9	35.55	5.60
Limed									
Check	19.5 20.1	19.80	24.9 19.2	22.05	2.0 6.9	4.45	46.4 46.2	46.30	
Ammonium phosphate	27.0 26.7	26.85	11.8 10.3	11.05	4.0 3.8	3.90	42.8 40.8	41.80	−4.50
Acid phosphate	25.0 24.5	24.75	17.0 20.2	18.60	2.0 1.8	1.90	44.0 46.5	45.25	−1.05
Basic slag	26.4 21.0	23.70	15.7 23.0	19.35	2.3 1.6	1.95	44.4 45.6	45.00	−1.30
Raw rock phosphate	28.5 20.7	24.60	21.0 19.7	20.35	4.7 1.8	3.25	54.2 42.2	48.20	1.90
Sassafras loam (86.1 mgm. of P_2O_5 added)									
Unlimed									
Check	6.0 11.3	8.65	5.0 3.5	4.25	2.2 1.9	2.05	13.2 16.7	14.95	
Ammonium phosphate	12.7 16.3	14.54	6.0 4.7	5.35	2.1 1.9	2.00	20.8 22.9	21.85	6.90
Acid phosphate	18.0 17.0	17.50	6.0 5.9	5.95	1.7 1.8	1.75	25.7 24.7	25.20	10.25
Basic slag	5.5 14.0	9.75	8.4 5.4	6.90	2.6 1.5	2.05	16.5 20.9	18.70	3.75
Raw rock phosphate	14.0 9.6	11.80	4.0 4.0	4.00	1.3 2.1	1.70	19.3 15.7	17.50	2.55

TABLE 18—(Continued)

TREATMENT	FIRST CROP		SECOND CROP		THIRD CROP		AVERAGE		
	Weight	Average	Weight	Average	Weight	Average	Weight	Average	Increase over check
Limed									
	grams	*grams*	*grams*	*grams*	*grams*	*grams*	*grams*	*grams*	*grams*
Check	13.8 12.4	13.10	9.5 7.1	8.30	6.9 6.5	6.70	30.2 26.0	28.10	
Ammonium phosphate	15.0 19.2	17.0	14.5 14.5	14.50	8.1 6.9	7.50	37.6 40.6	39.10	11.00
Acid phosphate	16.0 16.5	16.25	15.4 15.1	15.25	9.1 7.1	8.10	40.5 38.7	39.60	11.50
Basic slag	16.0 11.2	13.60	17.7 23.4	20.55	6.7 6.9	6.80	40.4 41.5	40.95	12.85
Raw rock phosphate	10.4 7.8	9.10	16.0 15.6	15.80	7.5 7.3	7.40	33.9 30.7	32.30	4.20
Norfolk sand (172.2 mgm. of P_2O_5 added)									
Unlimed									
Check	23.6 22.2	22.90	8.5 3.1	5.80	1.3 1.2	1.25	33.4 26.5	29.95	
Ammonium phosphate	26.8 21.7	24.25	6.5 13.1	9.80	1.5 1.1	1.30	34.8 35.9	35.35	5.40
Acid phosphate	23.6 21.0	22.30	10.6 9.0	9.80	1.8 1.0	1.40	36.0 31.0	33.50	3.55
Basic slag	21.4 21.3	21.35	5.7 13.6	9.65	1.5 1.5	1.50	28.6 36.4	32.50	2.55
Raw rock phosphate	19.5 16.5	18.00	16.8 13.0	9.90	2.5 2.5	2.50	38.8 32.0	35.4	5.45
Limed									
Check	19.5 20.1	19.80	24.9 19.2	22.05	2.0 6.9	4.45	46.4 46.2	46.30	
Ammonium phosphate	18.4 22.5	20.45	25.3 22.0	23.65	2.2 2.5	2.35	45.9 47.0	46.45	0.15
Acid phosphate	23.2 15.4	19.30	18.9 25.7	22.30	5.3 3.9	4.60	47.4 45.0	46.20	−0.10
Basic slag	22.5 18.5	20.50	24.5 23.9	24.20	2.5 2.2	2.35	49.5 44.6	47.05	0.75
Raw rock phosphate	16.0 14.2	15.10	21.2 25.7	23.45	3.0 6.0	4.50	40.2 45.9	43.05	−3.25

TABLE 18—(Concluded)

TREATMENT	FIRST CROP		SECOND CROP		THIRD CROP		AVERAGE		
	Weight	Average	Weight	Average	Weight	Average	Weight	Average	Increase over check
Sassafras loam (172.2 mgm. of P_2O_5 added)									
Unlimed									
	grams	*grams*	*grams*	*grams*	*grams*	*grams*	*grams*	*grams*	*grams*
Check	6.0		5.0		2.2		13.2		
	11.3	8.65	3.5	4.25	1.9	2.05	16.7	14.95	
Ammonium phosphate	10.0		15.4		1.7		27.1		
	9.8	9.90	9.4	12.40	1.3	1.50	20.5	23.8	8.85
Acid phosphate	20.7		5.5		2.6		28.8		
	16.8	18.75	8.9	7.20	1.4	2.00	27.1	27.95	13.00
Basic slag	9.6		13.0		1.3		23.9		
	13.9	11.25	12.0	12.50	1.9	1.60	27.8	25.85	10.90
Raw rock phosphate	15.0		3.7		1.2		19.9		
	10.6	12.80	5.8	4.75	1.5	1.35	17.9	18.9	3.95
Limed									
Check	13.8		9.5		6.9		30.2		
	12.4	13.10	7.1	8.30	6.5	6.70	26.0	28.10	
Ammonium phosphate	14.1		17.7		8.2		40.0		
	14.3	14.20	23.9	20.80	6.5	7.35	44.7	42.35	14.25
Acid phosphate	14.8		18.6		5.7		39.1		
	14.0	14.40	23.2	20.90	5.8	5.75	43.0	41.05	12.95
Basic slag	10.0		17.0		8.2		35.2		
	12.6	11.30	20.4	18.70	5.9	7.05	38.9	37.05	8.95
Raw rock phosphate	9.8		14.1		6.8		30.7		
	10.5	10.15	14.2	14.15	8.0	7.40	32.7	31.70	3.60

other two fertilizers third and fourth. With larger amounts of phosphorus present the order of availability was acid phosphate, basic slag, ammonium phosphate and raw rock phosphate. As previously stated, the high acidity in these pots greatly injured the plants. The results indicate that acid phosphate, as has been previously shown, does not increase the acidity and hence gives comparatively good results in this acid soil. Basic slag is also beneficial for the same reason. Ammonium phosphate has been shown to increase the acidity somewhat and hence shows a lower availability than acid phosphate.

On the other hand, in the presence of lime the availability of ammonium phosphate and acid phosphate was practically the same with both rates of application. In one case basic slag was slightly superior to all the other fertilizers, and in the other it was not quite as good as ammonium phosphate or acid phosphate. The sample of basic slag used in this experiment had been used in previous pot experiments at the New Jersey Agricultural Experi-

ment Station and had shown exceptionally good results. No doubt the ordinary commercial basic slag would have shown lower results than are here recorded. Raw rock phosphate proved to be relatively unavailable, as has been repeatedly shown.

In general, this experiment indicates that on the basis of the phosphorous present, ammonium phosphate is of the same availability as acid phosphate. If the soil is acid and low in phosphorus the addition of ground limestone will probably increase the availability of ammonium phosphate, but if the soil is acid and high in phosphorus the availability may be actually decreased, or apparently so, by making conditions favorable for the utilization of the phosphorus already in the soil rather than that added.

TABLE 19

Availability of phosphate fertilizers in quartz sand

TREATMENT	FIRST CROP WEIGHT	SECOND CROP WEIGHT	TOTAL WEIGHT	AVERAGE	INCREASE OVER CHECK
	grams	*grams*	*grams*	*grams*	*grams*
Check	2.8	2.1	4.9		
	3.0	2.6	5.6	5.25	
Ammonium phosphate	13.0	2.5	15.5		
	12.4	2.5	14.9	15.20	9.95
Acid phosphate	13.0	2.0	15.0		
	12.0	2.5	14.5	14.75	9.50
Raw rock phosphate	5.0	3.3	8.3		
	5.5	2.9	8.4	8.35	3.10

Comparative availability of various phosphate fertilizers in quartz sand under greenhouse conditions

For this experiment 10-pound portions of quartz sand were weighed out into pots and 1 gm. of K_2SO_4, 0.3 gm. of $MgSO_4$, 0.15 gm. of $Fe_2(SO_4)_3$ and 5 gm. of $CaCO_3$ added to each in addition to phosphorus in the form of ammonium phosphate, acid phosphate or raw rock phosphate. In each case the amount of phosphorus present was equivalent to that in 2 gm. of acid phosphate. The acid phosphate analyzed 15 per cent P_2O_5 and the raw rock phosphate 31.41 per cent. All pots except those containing ammonium phosphate received 1.3 gm. of ammonium sulfate; in the latter case allowance was made for the nitrogen present in the fertilizer. Two crops of buckwheat were grown following the one application of fertilizers. The dry weights of the plants are given in table 19.

From the results of the experiment in quartz sand we may say that there is practically no difference between the fertilizing value of phosphorus in ammonium phosphate and that in acid phosphate. The slight difference

noted here is in favor of ammonium phosphate. Raw rock phosphate is about one-third as efficient as the other two fertilizers.

GERMINATION TESTS

While the experiments reported above had not indicated that ammonium phosphate would injure the growth of young plants it seemed desirable to make some direct tests to determine what crops are most easily injured by high concentrations of the salt; also how much can be safely applied to various soil types; the relative toxicity as compared with other common nitrogenous fertilizers; and finally, methods of application and general treatment that will lessen the injurious effect of a given application or enable larger applications to be made safely. Knowing the composition and method of preparation of the commercial ammonium phosphate it did not seem likely that it would

TABLE 20

Effect of ammonium phosphate on germination in a loam soil. Percentage germination

AMMONIUM PHOSPHATE APPLIED	WHEAT	COWPEAS	BUCKWHEAT	RAPE	BARLEY	OATS	CORN	VETCH
grams								
0.000	78.5	100.0	100.0	100.0	100.0	100.0	100.0	80.0
0.025	71.4	75.0	100.0	85.7	100.0	85.7	100.0	50.0
0.050	64.3	62.5	100.0	92.9	85.7	100.0	100.0	60.0
0.100	85.7	87.5	90.0	71.4	100.0	78.5	100.0	60.0
0.250	78.5	50.0	100.0	100.0	92.9	100.0	100.0	50.0
0.500	71.4	50.0	100.0	92.9	92.9	100.0	100.0	70.0
1.000	78.5	50.0	100.0	85.7	85.7	100.0	100.0	70.0
2.500	92.9	50.0	90.0	64.3	100.0	100.0	100.0	70.0
5.000	64.3	0.0	100.0	7.1	85.7	92.9	100.0	10.0

produce any greater injury than the pure mono-ammonium phosphate salt. That this assumption was found to be correct is shown by data given later, therefore, even though the commercial ammonium phosphate was used throughout the germination tests, it may be safely assumed that practically the same results would have been obtained with the pure salt.

The method adopted in the experimental work was to weigh out into tumblers 200 gm. of soil, in some cases moist and in other cases air-dried. The fertilizer was either mixed with the entire 200 gm. of soil or applied in a layer at some portion of the soil mass, frequently in direct contact with the seeds. The experiments were carried out at room temperature and at optimum moisture content for the various soils used. The details of the procedure used for each of the different experiments are given as these are discussed. The germination tests were always carried out in duplicate, with the exception of the first experiment, but for the purpose of conserving space only the averages are given in the following tables.

Effect of ammonium phosphate in various concentrations on different crops in a loam soil

This experiment was intended to serve merely as a preliminary trial to determine the approximate concentration required to injure the germination of various seeds. Moist loam soil was obtained directly from the field and weighed out in 200-gm. portions into tumblers. The fertilizer was mixed with the entire sample of soil. In the case of corn and cowpeas eight seeds were planted per tumbler, ten of buckwheat and vetch and fourteen of all the remainder. The results are given in table 20 in terms of percentage germination.

It will be noticed that the results are somewhat erratic, in many cases due to lack of duplicate tumblers for each rate of application, the small number of seeds used; and in some cases the poor vitality of the seeds. The results as a whole, however, do establish in a general way the point at which toxic effects on germination begin to show. These toxic points, as shown by the figures, agreed rather closely with the actual appearance of the plants.

When the plants were older some of them began to die, even in concentrations below 1000 mgm. At the end of three weeks the cowpeas were dying in the 500-mgm. tumbler, buckwheat in the 250-mgm. tumbler, and corn in the 500-mgm. tumbler. There is but little doubt that the greatest injury of the fertilizer applied under field conditions would not be to germination, itself, but rather to the young plants after they are a few inches in height.

Effect of ammonium phosphate, ammonium sulfate, and acid phosphate on corn, wheat, and soybeans in a loam soil

This experiment was carried out in the same manner as the one previously discussed except that an air-dried Penn loam soil was used and 12 seeds planted in each tumbler. The fertilizer was thoroughly mixed with the entire 200 gm. of soil. The corn was cut 30 days and the wheat and soybeans 40 days, after the time of planting, and the weight taken. In the case of ammonium sulfate the same amount of nitrogen was applied as in the ammonium phosphate tumblers. Likewise, the acid phosphate applications were based on the amount of phosphoric acid in the ammonium phosphate. In another series of tumblers the acid phosphate and ammonium sulfate were combined, giving a mixture with the same amount of nitrogen and phosphorus as is present in ammonium phosphate. The summarized results are given in table 21, and a portion of the data shown diagrammatically in figure 8.

A glance at the figures shows that the diminution in the yield of green matter was rather gradual as the applications of the two fertilizers, ammonium phosphate and ammonium sulfate, increased. At the lower rates of application there was a stimulation in the growth of corn and wheat due to the plant-food added, and then at higher concentrations the decline was gradual, many

of the plants having died before harvesting. Acid phosphate showed a marked stimulation up to amounts equivalent to 4 gm. of ammonium phosphate per tumbler, and then a sudden drop. Where acid phosphate and ammonium

TABLE 21

Effect of fertilizers on the germination and early growth of corn, wheat and soybeans in a loam soil

TREATMENT (GM.)	AVERAGE GERMINATION			AVERAGE WEIGHT		
	Corn	Wheat	Soy-beans	Corn	Wheat	Soy-beans
	per cent	*per cent*	*per cent*	*grams*	*grams*	*grams*
Check	100.0	95.8	95.8	6.9	0.58	11.5
Ammonium phosphate 0.250	100.0	95.8	100.0	7.5	0.89	9.6
Ammonium phosphate 0.500	100.0	83.3	95.8	8.6	0.90	9.1
Ammonium phosphate 1.000	100.0	91.7	54.2	7.3	0.94	2.3
Ammonium phosphate 2.000	100.0	87.5	95.8	5.2	0.92	3.2
Ammonium phosphate 3.000	95.8	66.6	33.3	4.6	0.60	0.2
Ammonium phosphate 4.000	100.0	91.7	45.8	2.8	0.44	0.0
Ammonium phosphate 5.000	95.8	83.3	45.8	2.8	0.24	0.0
Ammonium sulfate 0.131	100.0	95.8	95.8	9.0	1.00	10.5
Ammonium sulfate 0.263	100.0	95.8	100.0	8.7	0.95	9.3
Ammonium sulfate 0.526	100.0	91.7	95.8	7.6	0.91	8.7
Ammonium sulfate 1.051	100.0	100.0	83.3	6.0	1.02	6.6
Ammonium sulfate 1.577	100.0	75.0	37.5	5.6	0.63	1.1
Ammonium sulfate 2.102	95.8	79.2	37.5	3.2	0.46	0.0
Ammonium sulfate 2.628	100.0	62.5	25.0	2.4	0.14	0.0
Ammonium sulfate 0.131+acid phosphate 0.719	100.0	100.0	91.7	8.9	1.24	8.1
Ammonium sulfate 0.263+acid phosphate 1.438	100.0	91.7	83.3	7.5	0.88	8.3
Ammonium sulfate 0.526+acid phosphate 2.875	100.0	87.5	100.0	6.8	0.80	9.2
Ammonium sulfate 1.051+acid phosphate 5.750	95.8	95.8	83.3	4.6	0.93	3.6
Ammonium sulfate 1.577+acid phosphate 8.625	100.0	100.0	45.8	4.3	0.38	0.0
Ammonium sulfate 2.102+acid phosphate 11.501	95.8	100.0	41.6	2.7	0.26	0.0
Ammonium sulfate 2.628+acid phosphate 14.386	95.8	87.5	25.0	1.7	0.15	0.0
Acid phosphate 0.719	100.0	100.0	87.5	7.4	1.13	8.2
Acid phosphate 1.438	100.0	95.8	83.3	8.1	1.19	8.5
Acid phosphate 2.875	100.0	91.7	79.2	7.6	1.03	9.6
Acid phosphate 5.750	100.0	79.2	91.7	7.4	1.07	8.6
Acid phosphate 8.625	100.0	95.8	83.3	6.4	1.18	7.8
Acid phosphate 11.501	25.0	16.6	8.3	0.2	0.00	0.0
Acid phosphate 14.386	0.0	8.3	4.2	0.0	0.00	0.0

sulfate were used together the growth more nearly corresponded to that of ammonium phosphate or ammonium sulfate when used alone. The results with soybeans showed the same trend, except as already stated, the fertilizers were much more toxic and showed less tendency to stimulate. In general,

ammonium phosphate is slightly less toxic than ammonium sulfate used in combination with acid phosphate for corn and wheat and more

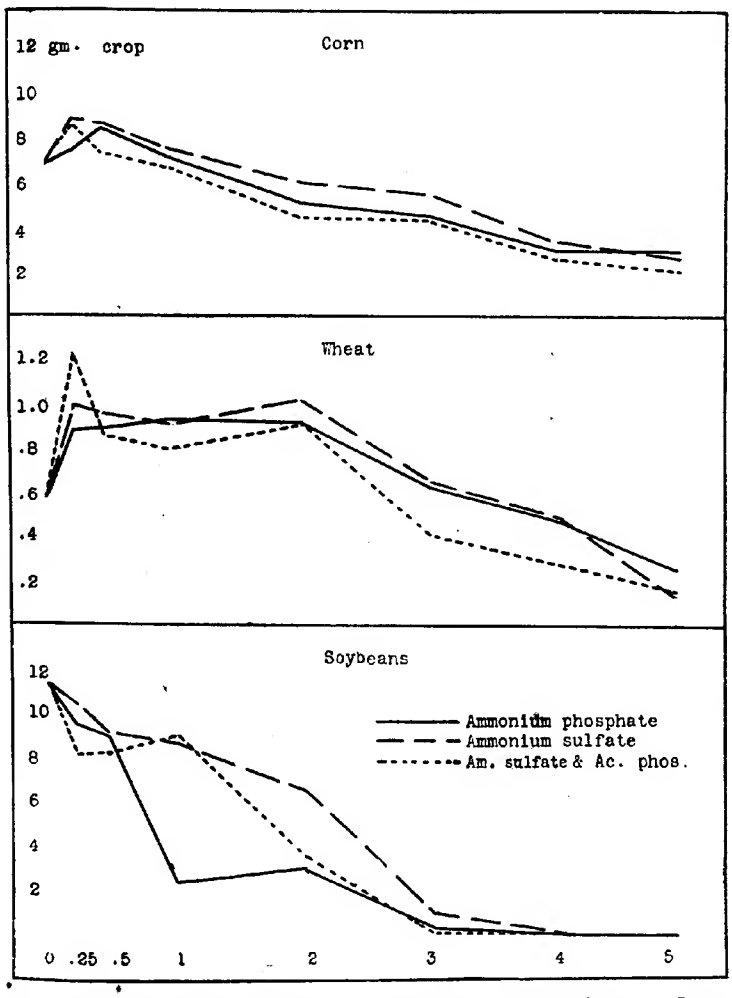

FIG. 8. DIAGRAM SHOWING THE EFFECT OF FERTILIZERS ON THE EARLY GROWTH OF PLANTS IN A LOAM SOIL

(Figures represent grams of ammonium phosphate. Ammonium sulfate applications contained equivalent amounts of nitrogen.)

toxic than ammonium sulfate used singly. For soybeans ammonium phosphate was more toxic than ammonium sulfate alone or in combination with acid phosphate.

TABLE 22

Effect of various nitrogenous fertilizers, alone and in combination, on the germination and growth of corn in a limed and an unlimed loam soil

TREATMENT		0 MGM. NITROGEN	56 MGM. NITROGEN	111 MGM. NITROGEN	223 MGM. NITROGEN	334 MGM. NITROGEN	446 MGM. NITROGEN	557 MGM. NITROGEN
Limed								
Check	Germination (per cent)	100.0						
	Weight (gram)	6.3						
Ammonium phosphate	Germination		87.5	91.7	100.0	91.7	100.0	70.8
	Weight		5.6	6.3	5.0	8.3	6.7	3.5
Ammonium sulfate	Germination		83.3	79.2	87.5	91.7	91.7	58.3
	Weight		5.7	5.4	4.7	5.2	3.5	9.9
Ammonium sulfate + acid phosphate	Germination		87.5	100.0	100.0	100.0	100.0	87.5
	Weight		5.4	6.4	6.6	8.1	6.5	3.0
Sodium nitrate + acid phosphate	Germination		100.0	87.5	87.5	75.0	66.6	4.2
	Weight		7.2	6.1	4.1	1.2	1.5	0.0
Unlimed								
Check	Germination	87.5						
	Weight	7.3						
Ammonium phosphate	Germination		91.7	87.5	95.8	91.7	100.0	91.7
	Weight		10.0	9.2	3.9	3.0	2.4	0.8
Ammonium sulfate	Germination		91.7	79.2	87.5	91.7	91.7	66.6
	Weight		10.8	7.9	3.6	3.0	1.0	1.2
Ammonium sulfate + acid phosphate	Germination		100.0	91.7	91.7	91.7	100.0	87.5
	Weight		10.1	8.7	3.3	2.6	1.2	1.8
Sodium nitrate + acid phosphate	Germination		100.0	100.0	91.7	70.8	50.0	20.8
	Weight		11.5	8.6	4.5	0.9	0.5	0.0

		0 MGM. NITROGEN	111 MGM. NITROGEN	223 MGM. NITROGEN	446 MGM. NITROGEN	670 MGM. NITROGEN	893 MGM. NITROGEN	1115 MGM. NITROGEN
Limed								
Check	Germination	100.0						
	Weight	6.3						
Ammonium phosphate + ammonium sulfate	Germination		91.7	95.8	91.7	95.8	58.3	41.6
	Weight		7.0	5.3	5.5	3.6	0.4	0.5
Ammonium phosphate + sodium nitrate	Germination		95.8	79.2	83.3	75.0	54.2	0.0
	Weight		7.3	2.9	2.6	0.5	0.2	0.0
Ammonium sulfate + acid phosphate	Germination		83.3	100.0	100.0	100.0	66.6	20.8
	Weight		5.0	5.8	5.7	2.5	0.5	0.0
Unlimed								
Check	Germination	87.5						
	Weight	7.3						
Ammonium phosphate + ammonium sulfate	Germination		91.7	95.8	95.8	87.5	58.3	20.8
	Weight		10.3	6.1	2.5	1.0	0.4	0.0
Ammonium phosphate + sodium nitrate	Germination		91.7	100.0	87.5	6.66	29.1	0.0
	Weight		9.0	8.2	2.8	0.5	0.1	0.0
Ammonium sulfate + acid phosphate	Germination		91.7	91.7	95.8	83.3	33.3	58.3
	Weight		7.3	4.1	2.4	0.6	0.3	0.4

Effect of various nitrogen fertilizers alone and in combination on germination and plant growth in a limed and an unlimed loam soil

Since the experiment reported in table 21 had indicated that perhaps the injurious effects of fertilizers at high concentrations when applied alone may

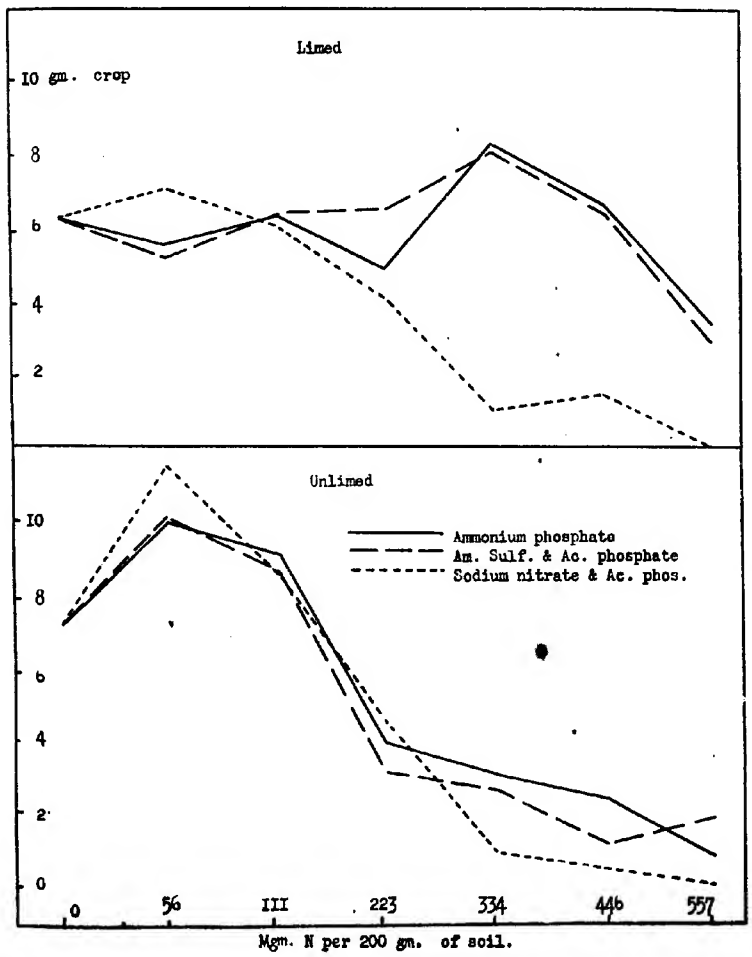

FIG. 9. DIAGRAM SHOWING THE EFFECT OF EXCESSIVE QUANTITIES OF NITROGENOUS FERTILIZERS ON THE EARLY GROWTH OF CORN

be greater than when used in combinations, the experiment reported below was planned. Half of the tumblers received 3 gm. of ground limestone each, while the remainder were left unlimed. Ten seeds were planted in each tumbler after mixing the fertilizer with the 200 gm. of soil. The yield of the corn

crop as well as the percentage of germination is given in table 22, and shown diagrammatically in figures 9 and 10.

From the data presented in table 22 it may be stated that on limed soil ammonium phosphate and the equivalent mixture of acid phosphate and

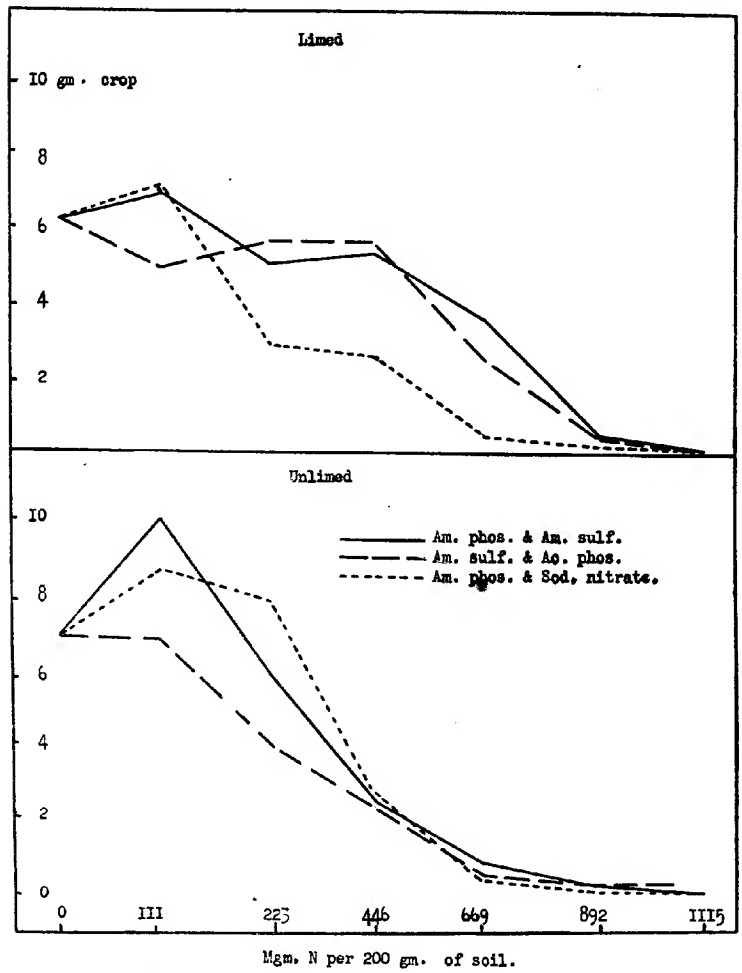

FIG. 10. DIAGRAM SHOWING THE EFFECT OF VARIOUS COMBINATIONS OF NITROGENOUS FERTILIZERS ON THE EARLY GROWTH OF CORN

ammonium sulfate produced equal effects. There was no great difference in the results where ammonium sulfate was used alone or in combination with acid phosphate. On unlimed soil ammonium phosphate was slightly less toxic than the equivalent mixture of ammonium sulfate and acid phosphate.

On both limed and unlimed soil ammonium phosphate in combination with ammonium sulfate was less toxic than the same amount of nitrogen and phosphorus in the form of ammonium sulfate and acid phosphate. On limed soil sodium nitrate was more toxic than either ammonium phosphate or

TABLE 23

Relation of type of soil to the injurious effects of fertilizers on the germination of corn

TREATMENT		0 MGM. NITROGEN	28 MGM. NITROGEN	56 MGM. NITROGEN	112 MGM. NITROGEN	223 MGM. NITROGEN	335 MGM. NITROGEN	446 MGM. NITROGEN	558 MGM. NITROGEN
Clay									
Check	Germination (per cent)	95.8							
	Weight (gram)	6.6							
Ammonium phosphate	Germination		100.0	95.8	91.7	100.0	87.5	100.0	87.5
	Weight		6.9	6.9	7.0	4.2	2.8	3.2	2.3
Ammonium sulfate	Germination		95.8	91.7	95.8	83.3	95.8	91.7	87.5
	Weight		6.1	6.2	4.8	2.4	2.3	1.7	1.0
Silt loam									
Check	Germination	95.8							
	Weight	7.0							
Ammonium phosphate	Germination		87.5	100.0	95.8	95.8	91.7	91.7	79.2
	Weight		8.3	7.2	6.7	6.7	5.5	2.9	1.9
Ammonium sulfate	Germination		95.8	91.7	95.8	91.7	100.0	91.7	87.5
	Weight		7.8	7.7	7.4	5.1	4.5	3.5	3.0
Sandy loam									
Check	Germination	95.8							
	Weight	7.2							
Ammonium phosphate	Germination		83.3	87.5	100.0	87.5	87.5	95.8	87.5
	Weight		4.8	4.2	3.0	0.9	0.5	0.0	0.0
Ammonium sulfate	Germination		95.8	95.8	100.0	91.7	91.7	29.1	54.2
	Weight		5.5	4.6	2.9	1.0	0.6	0.0	0.0
Sand									
Check	Germination	100.0							
	Weight	6.2							
Ammonium phosphate	Germination		91.7	95.8	91.7	33.3	12.4	4.2	0.0
	Weight		4.8	1.0	0.5	0.0	0.0	0.0	0.0
Ammonium sulfate	Germination		100.0	95.8	83.3	66.6	25.0	20.8	8.3
	Weight.		5.5	1.6	1.0	0.0	0.0	0.0	0.0

ammonium sulfate with acid phosphate. On unlimed soil sodium nitrate with acid phosphate was more toxic than ammonium phosphate, but ammonium phosphate plus sodium nitrate had practically the same effect as ammonium phosphate plus ammonium sulfate.

Relation of type of soil to the germination of corn in the presence of excessive quantities of fertilizers

The germination work reported in the preceding pages had shown, in a rough way, that the heavier the soil the larger is the amount of fertilizer required to affect germination. In order to have a direct comparison between soils, four types were chosen, namely, clay, silt loam, sandy loam and sand. This experiment was carried out in the same manner as those previously discussed, using 200 gm. of air-dried soil and planting 12 kernels of corn per tumbler. The counts were made on the ninth day and the crop harvested and weighed green on the sixteenth day. The results are given in table 23.

The results show very strikingly that very much larger applications of either ammonium phosphate or ammonium sulfate are required for the heavier soils than for the lighter ones to produce a marked injury. Applications which entirely prevented the growth of corn in the sandy soil produced only a slight injury in the clay and silt loam soils. It should be stated that the results obtained for the clay in comparison with the silt loam may be somewhat misleading. Clay, being the heavier soil, would be expected to require more fertilizer to produce an injury and this would probably be true for most clay soils, but the soil used in this experiment was very granular and perhaps we might say cloddy, so that even after sieving through a coarse sieve the soil was quite loose and behaved as a coarser soil. On the other hand, the silt loam was very fine and exceedingly compact when moistened.

In general, it will be noticed that the injury produced by a given amount of nitrogen as ammonium phosphate was about equal to that produced by a like amount of nitrogen as ammonium sulfate except in the clay soil where ammonium phosphate was less toxic than ammonium sulfate. In this experiment no acid phosphate equal to the amount of phosphoric acid in ammonium phosphate was applied to the ammonium sulfate tumblers.

Effect of ammonium phosphate and ammonium sulfate on the germination of corn in quartz sand and in a sandy soil

This experiment, carried out in the same manner as those previously discusssed, was planned to determine the maximum amount of fertilizers that may be applied to a pure sand and a very sandy soil without injury. The quartz sand, of course, need not be considered from the standpoint of field conditions, but the sandy soil was no lighter or poorer than might be used to grow crops in extreme cases. Such a soil would certainly be an exception, though, and represents the extreme that would ever be put under cultivation.

The amounts of ammonium sulfate and acid phosphate used were always equivalent to the amount of nitrogen and phosphoric acid present in the corresponding ammonium phosphate tumblers. Ten kernels of corn were planted in each tumbler, the germination counts being made on the tenth

day and the crop harvested at the end of 25 days. These figures are given in table 24.

It will be noticed that in the case of quartz sand there is an appreciable decrease in germination with 200 mgm. of ammonium phosphate or the corresponding amount of ammonium sulfate, and on the basis of the amount of nitrogen present there is little difference in the toxicity of the two fertilizers. In growth up until the end of the 25-day period it is clearly brought out that even the smallest application used, 50 mgm. per tumbler, produced some injury while 100 mgm. decreased the yield about one-half.

Considering the results with the sandy soil it will be noticed that the decrease in germination began at 750 mgm. of ammonium phosphate per tumbler

TABLE 24

Effect of fertilizers on the germination of corn in quartz sand and in sandy soil

TREATMENT		0 MGM. NITROGEN	5.6 MGM. NITROGEN	11.1 MGM. NITROGEN	22.3 MGM. NITROGEN	33.4 MGM. NITROGEN	44.6 MGM. NITROGEN	55.7 MGM. NITROGEN	83.5 MGM. NITROGEN	111.5 MGM. NITROGEN	167.2 MGM. NITROGEN	223.0 MGM. NITROGEN	278.7 MGM. NITROGEN
Quartz sand													
Check	Germination (per cent)	80.8											
	Weight (gram)	1.1											
Ammonium phosphate	Germination		83.3	83.3	58.3	29.1	29.1	4.2	4.2	0.0	0.0		
	Weight		0.8	0.4	0.1	0.0	0.0	0.0	0.0	0.0	0.0		
Ammonium sulfate	Germination		79.2	75.0	66.6	16.6	16.6	4.2	0.0	0.0	0.0		
	Weight		0.8	0.5	0.1	0.0	0.0	0.0	0.0	0.0	0.0		
Sandy soil													
Check	Germination	83.3											
	Weight	1.9											
Ammonium phosphate	Germination		79.2	83.3	83.3		83.3		54.2	33.3	4.2	0.0	0.0
	Weight		2.0	2.3	2.3		0.6		0.1	0.0	0.0	0.0	0.0
Ammonium sulfate	Germination		75.0	83.3	83.3		79.2		45.8	16.6	4.2	4.2	0.0
	Weight		2.0	2.6	lost		0.5		0.1	0.0	0.0	0.0	0.0

or the corresponding amount of ammonium sulfate. Again the two fertilizers showed similar effects on germination. The decrease in growth began at about 200 or 300 mgm. of ammonium phosphate per tumbler.

Comparison of methods of application

This experiment was carried out with the purpose of determining if the injury to germination is lessened by placing the fertilizer at a short distance from the germinating seeds. A Norfolk sand was used. The data are given in table 25, the germination counts being made at the end of 13 days and the plants harvested on the twenty-first day.

The position of the fertilizer in relation to the seed again had very little

effect except possibly in the tumblers where the seeds were planted one inch deep and the fertilizer placed an inch below the seeds. More fertilizer was required in these tumblers to produce a given effect than in any of the other tumblers.

Effect of various nitrogenous salts on the germination of corn

The previous experiments had fixed rather definitely the relation of type of soil to fertilizer injury and also the relative injury that may be expected from the use of ammonium phosphate in comparison with ammonium sulfate or acid phosphate. However, since the data reported in this paper are primarily a study of an impure ammonium phosphate it is of interest to make a further comparison to determine if this commercial phosphate salt will behave in a similar manner to the pure salt, or whether the impurities present

TABLE 25

Comparison of method of application

AMMONIUM PHOSPHATE APPLIED	FERTILIZER IN CONTACT WITH SEED 1.5 INCHES DEEP		FERTILIZER 1 INCH DEEP; SEED 2 INCHES		FERTILIZER 2 INCHES DEEP; SEED 1 INCH		FERTILIZER UNIFORMLY DISTRIBUTED; SEED 1.5 INCHES DEEP	
	Average germination	Average weight	Average germination	Average weight	Average germination	Average weight	Average germination	Average weight
grams	*per cent*	*grams*	*per cent*	*grams*	*per cent*	*grams*	*per cent*	*grams*
Check	97.5	5.0						
0.10	100.0	5.5	100.0	5.6	100.0	6.1	100.0	5.9
0.20	100.0	5.3	100.0	4.6	100.0	6.3	100.0	5.1
0.40	100.0	2.8	100.0	3.5	100.0	4.0	85.0	1.6
0.80	90.0	0.9	95.0	0.8	95.0	3.2	90.0	0.8
1.60			25.0	0.1	100.0	1.7	0.0	0.0
2.40			0.0	0.0	95.0	0.6	0.0	0.0
3.20							0.0	0.0

increase or decrease the injury to germination and plant growth. Furthermore, what relation does ammonium phosphate bear to other nitrogenous salts, especially ammonium salts. The soil chosen for testing these points was the Sassafras loam used in previous experiments. Ten kernels of corn were planted per tumbler and the fertilizers placed in direct contact with the seeds. With the exception of ammonium nitrate the applications were based on the amount of nitrogen present. Only the ammonium nitrogen was considered in the case of this salt. The germination counts were made after 24 days and the crop harvested at the same time. The results are given in table 26.

From the standpoint of the crude ammonium phosphate the most interesting point brought out in the table is that with large applications of the fertilizer the toxic effects are usually no greater than with the pure mono-ammonium phosphate, proving that the effects produced are not to any great extent, if

at all, due to the method of making the fertilizer or to any impurities present. The di- and tri-ammonium phosphates were somewhat less toxic on the basis of the amount of nitrogen present than was the mono form. In agreement with previous work the injury produced by a given amount of the ammonium phosphate fertilizer was practically the same as with ammonium sulfate. Ammonium chloride was the most toxic of all the salts used, undoubtedly because of the chloride radical. Ammonium nitrate, on the basis of the amount of ammonia present, showed about the same degree of toxicity as the ammonium phosphate fertilizer, ammonium sulfate or mono-ammonium phosphate, regardless of the fact that twice as much nitrogen was present,

TABLE 26

Effect of nitrogenous salts on the germination of corn

TREATMENT		0 MGM. NITROGEN	11.5 MGM. NITROGEN	23.0 MGM. NITROGEN	46.0 MGM. NITROGEN	80.5 MGM. NITROGEN	115.0 MGM. NITROGEN	172.0 MGM. NITROGEN
Check	Germination (per cent)	86.6						
	Weight (grams)	5.1						
"Ammo Phos"	Germination		85.0	95.0	90.0	70.0	65.0	25.0
	Weight		5.3	7.0	3.0	2.9	0.8	0.1
$NH_4H_2PO_4$	Germination		95.0	90.0	90.0	65.0	40.0	40.0
	Weight		4.5	5.0	4.1	2.6	0.7	0.3
$(NH_4)_2HPO_4$	Germination		80.0	100.0	100.0	90.0	65.0	25.0
	Weight		4.3	7.2	7.4	3.2	1.8	0.2
$(NH_4)_3PO_4$	Germination		100.0	95.0	100.0	95.0	75.0	70.0
	Weight		6.5	5.2	5.8	3.6	2.5	2.4
$(NH_4)_2SO_4$	Germination		100.0	95.0	100.0	90.0	90.0	60.0
	Weight		5.8	7.0	5.4	3.2	2.2	0.3
NH_4Cl	Germination		80.0	80.0	85.0	55.0	40.0	15.0
	Weight		5.0	5.1	1.3	0.6	0.2	0.0
NH_4NO_3	Germination		90.0	90.0	85.0	75.0	60.0	10.0
	Weight		6.7	6.7	3.3	2.8	0.9	0.0
$NaNO_3$	Germination		85.0	90.0	90.0	85.0	80.0	20.0
	Weight		5.8	6.9	5.4	2.2	1.9	0.1

half as ammonia and half as nitrate nitrogen. Sodium nitrate showed a slightly higher degree of toxicity than did the commercial ammonium phosphate fertilizer, but was not as toxic as ammonium chloride.

The results of this experiment indicate that the concentration of the salt, whatever that salt may be, is the primary cause of the injury to germination and young plants. Secondary to this is the acid radical and the base to which it is combined. In the case of ammonium salts, as is brought out in table 26, the presence of the chloride radical resulted in a much greater injury than the nitrate.

Germination of corn in the field in the presence of ammonium phosphate

After completing the germination tests in tumblers reported in the previous pages it seemed desirable to carry on some field work to determine just how much ammonium phosphate fertilizer may be applied to corn in the row without injury. Consequently on May 30, 1917 an experiment was begun on a Sassafras loam, which is the same soil used frequently in the germination experiments in the laboratory. Rows were laid off 1 foot apart and 50 kernels of corn planted per row, about 2 to 3 inches apart. The ammonium phosphate was applied in the row in a strip about 1 inch wide in direct contact with the seed, exactly as if applied with a seed drill having a fertilizer attachment. The rates of application were based on rows 32 inches apart. In other words, each linear foot received the same amount of ammonium phosphate as it

TABLE 27

Germination of corn in field fertilized with ammonium phosphate

POUNDS OF AMMONIUM PHOSPHATE PER ACRE IN ROW WITH SEED	GERMINATION (NUMBER OF KERNELS SPROUTED)							
	12 days				22 days			
	1	2	3	Average	1	2	3	Average
0	48	50	49	49	48	49	49	49
25	49	49	49	49	48	49	49	49
50	44	48	49	47	43	48	49	47
100	47	50	48	48	46	50	48	48
150	37	44	46	42	41	46	46	44
200	34	41	39	38	37	42	41	40
300	23	29	20	24	26	35	25	29
400	17	22	19	19	21	25	24	23
500	7	11	8	9	11	13	17	14

would in the field in rows 32 inches apart at the given rates per acre. The germination was counted on June 11 and 21 and is given in table 27.

The corn on the rows with 50 and 100 pounds appeared to be best and was equal in size, color and apparent vigor. The rows with 150 pounds showed the effects of too much fertilizer. The decrease in the size of the plants with increasing applications of fertilizer was about proportional to the numbers given for germination. With 150 pounds and over there were a few plants, about 5 to 10 per cent, which looked yellow and stunted, and will probably die. The balance were dark green and not injured to any extent, if at all. The corn on the rows with 50 and 100 pounds was about 9 inches high and of very good color at the time of making the second germination counts. In this test 100 pounds is a perfectly safe application per acre. This was a very wet season, however, which should be more favorable than a dry season. It should also be borne in mind that the soil used is fairly heavy. Further tests are necessary before general conclusions can be drawn.

General discussion of germination experiments

The results of the germination experiments show that ammonium phosphate when applied to the soil has practically the same effect upon germination as a mixture of acid phosphate and ammonium sulfate in the same proportion as the plant-food constituents present in the ammonium phosphate fertilizer. In fact, such a large amount of acid phosphate was required to injure germination that we may say that the ammonium phosphate produces results quite similar to ammonium sulfate used alone. It has also been brought out that any injury produced by the commercial ammonium phosphate is due to the concentration of the soluble salts and not to any impurities that are present, for the injury produced by chemically pure mono-ammonium phosphate is just as great. Other salts, such as ammonium chloride and sodium nitrate, may produce even greater injury than ammonium phosphate. Evidently, then, the acid radical exerts its influence, but with most salts it is the concentration of the soluble material about the germinating seeds that plays the primary rôle. As was brought out earlier it is very significant that ammonium nitrate, on the basis of the amount of ammonia present, showed no greater toxicity than ammonium phosphate or ammonium sulfate, yet there was twice as much nitrogen present, half as ammonia and the remainder as nitrate nitrogen.

Regarding the migration of soluble fertilizer salts in the soil, these experiments have shown that under the conditions prevailing in the tumblers the salts are carried from one point to another with sufficient rapidity to affect the germinating seed regardless of the method of application, whether at some particular point in the soil or uniformly distributed throughout the 200-gm. sample. However, these results must not be applied too closely to field conditions, where rains usually come at intermittent periods rather than as a small amount every day, as was the case in the tumblers.

Investigators disagree as to the rapidity with which soluble salts diffuse through the soil. Harris (9) shows that salts are transferred through the soil readily by moving water. Malpeaux and Lefort (23) showed that the diffusion of nitrates was comparatively slow and was practically the same laterally as vertically. Sprinkling accelerated downward as well as lateral diffusion but there was rather a rapid return to the upper layers of soil by capillarity. The diffusion was more rapid in the sandy soil but less uniform. He concludes that under ordinary conditions of rainfall there is a comparatively rapid diffusion of the nitrates in the surface soil. Müntz and Gaudeschon (25) conclude that the most soluble substances diffuse in the soil only with extreme slowness. The writer is of the opinion that under ordinary field conditions soluble salts are washed down to the subsoil and again brought to the surface by capillarity very readily, but that the lateral diffusion is almost negligible.

How much fertilizer may be applied safely under field conditions without

injury if in direct contact with the seed? This question has not been completely answered by the experiments reported but some progress has been made. In the first place the crop grown must be considered. Basing our statements on table 20 we may say that corn, buckwheat, oats, wheat and barley are resistant crops, while vetch, rape and cowpeas may be considered as relatively easily injured. Since most of this work deals with the germination of corn we should bear in mind that this crop is relatively resistant and a greater injury would be expected with some other crops, especially those with high oil content, such as cowpeas, soybeans and cotton.

Kearney and Harter (12), using alkali salts and eight species of plants, found that in water culture maize was on the whole the most resistant to pure solutions and cotton the least. Harris (8) working with alkali salts, found that the relative resistance of various crops in the seedling stage is usually in the following order, barley being the most resistant, barley, oats, wheat, alfalfa, sugar beets, corn and Canada field peas. Only about half as much alkali was required to prohibit the growth of crops in sand as in loam. He further states that the period of germination of seeds is considerably lengthened by the presence of soluble salts in the soil. The same observation was made by the writer, using the different fertilizers.

Another point which must be considered is the type of soil. As was brought out in table 23 the amount of fertilizer required to produce a given injury in a clay or silt loam soil is probably ten times as much as is required in a sandy soil, at least under laboratory conditions. The injury would probably not be quite as great under field conditions but nevertheless it is a well known fact that there is a big difference between soils. This may be partially attributed to the variations in the water-holding capacity, but also to the fact that the heavier soils have a greater absorbing capacity. On the other hand, it should be borne in mind that the fertilizer will distribute itself more rapidly in a lighter soil, and when applied in the row this factor would enter in to decrease the injury that we would otherwise expect to occur.

Judging from the one field experiment reported it would seem that it is reasonably safe to apply as much as 100 pounds of ammonium phosphate per acre drilled in the corn row on the average soil, but it must be borne in mind that this experiment was conducted during a wet month. It is to be regretted that lack of time prevented the carrying out of a variety of field experiments. The tumbler experiments, however, have been sufficient practically to prove that a given amount of nitrogen as sodium nitrate or ammonium sulfate will produce just as great an injury as the same amount of nitrogen as ammonium phosphate, but this does not settle the question because there is little experimental evidence to show how much of these salts may be applied with safety, in drills. It is usually customary to consider that such a fertilizer as ammonium sulfate in moderate amounts may be drilled in the row with safety, but in the light of the results reported here it seems doubtful. It is a well known fact that the farmer has a prejudice in favor of organic fertilizers even though

the nitrogen is not as available as that in ammonium sulfate. Perhaps this prejudice is after all a just one and it would seem so since it has grown up as a result of many years of experience. Even in so-called moderate amounts the ammoniate fertilizers, when applied in the drill, may produce marked injury, especially in dry seasons on sandy soils. The remedy seems to be either to apply smaller amounts of the soluble fertilizer in the drill, or to spread it over a wider strip in the row, or to make applications alongside the row or broadcast.

CONCLUSIONS

From the results of the work which has been reported in the preceding pages it may be briefly said that the ammonium phosphate fertilizer is in general of the same value as an equivalent amount of nitrogen as ammonium sulfate and phosphorus as acid phosphate, and may be used as a substitute for these other forms of plant-food. While in isolated cases this statement did not hold true, they were so few and under such abnormal soil conditions that they scarcely need be considered. In fact, the number of cases where ammonium phosphate was slightly superior to ammonium sulfate was larger than where the reverse was true. Ammonium phosphate is readily nitrified and utilized by both microorganisms and plants.

From the standpoint of injurious effects when applied in high concentrations, ammonium phosphate is again similar to ammonium sulfate and slightly less toxic than sodium nitrate. In its application in drills caution should be used in regard to the amount applied, but this statement applies to other soluble salts as well.

SUMMARY

The chief points which have been brought out in this thesis may be summarized as follows:

1. The commercial ammonium phosphate fertilizer used contains approximately 13.5 per cent of ammonia and 43 per cent of phosphoric acid, 96.5 per cent of which is either water- or citrate-soluble.

2. Ammonium sulfate and ammonium phosphate nitrify at approximately the same rate, while dried blood, cottonseed meal and tankage are considerably less available, usually in the order named, tankage being the least available.

3. A gradual increase in nitrate accumulation was found in tumblers up until the sixth week in a rich garden soil and until 8 to 10 weeks in a meadow soil. After the maximum accumulation in the garden soil the decline was quite rapid, showing the importance that may be attached to nitrate assimilation by microörganisms.

4. Calcium carbonate proved to be especially favorable for nitrification while calcium oxide sometimes caused an actual depression in the nitrification of ammonium sulfate and ammonium phosphate.

5. Ammonium phosphate increased the rate of ammonification of cotton-

seed meal but decreased the ammonia production from dried blood. Green alfalfa was not appreciably affected by the presence of the fertilizer.

6. Soil fungi were found to utilize nitrogenous salts in solution in the following order, sodium nitrate showing the poorest results, ammonium phosphate ammonium carbonate, ammonium sulfate, urea, ammonium nitrate and sodium nitrate.

7. As an average of all conditions the recoveries of nitrogen applied to Norfolk sand, yielding six crops—one of barley, four of buckwheat and one of corn, were 65.88, 61.10, 43.74 and 41.19, respectively, for ammonium phosphate, ammonium sulfate, dried blood and cottonseed meal. On a Sassafras loam the average recoveries in the same order were, 48.46, 50.42, 42.51 and 36.49. Liming usually increased the recovery of nitrogen. The crop yields were about in the same ratio as the nitrogen recoveries, except that under very acid conditions frequently ammonium sulfate showed a higher recovery of nitrogen than ammonium phosphate but a much smaller crop. The comparatively low recovery of the nitrogen added leads the writer to believe that a considerable amount of nitrogen escapes from soil as free nitrogen gas or ammonia or is given off from the plant itself.

8. The average of the results for two soils, limed and unlimed, with two rates of application of fertilizers, showed the following increases in lime requirement over checks, due to the fertilizers applied: ammonium sulfate, 794 pounds of CaO; ammonium phosphate, 525 pounds; dried blood, 263 pounds; and cottonseed meal, 113 pounds.

9. Pot experiments in sand showed the nitrogen in the commercial ammonium phosphate, the water-soluble extract of ammonium phosphate and sodium nitrate to be of practically the same availability. Water-insoluble ammonium phosphate showed only a slightly lower yield than the other forms of nitrogen.

10. Experiments in soils in the greenhouse, as an average, showed little difference in the availability of the phosphorus between ammonium phosphate, acid phosphate and basic slag, while raw rock phosphate gave much smaller increases. On very acid soils acid phosphate was sometimes superior to ammonium phosphate as a source of phosphorus, doubtless because the former does not increase the acidity to any great extent, if at all, while ammonium phosphate does increase the acidity somewhat. In quartz sand the relative increases over the check were for ammonium phosphate 9.95 gm., acid phosphate 9.50 gm. and raw rock phosphate 3.10 gm.

11. In germination experiments ammonium phosphate, on the basis of the amount of nitrogen present, showed about the same toxicity to germination and early growth as ammonium sulfate and less than sodium nitrate or ammonium chloride when applied to soils in high concentrations.

12. Very sandy soils require only about one-tenth as much fertilizer in tumblers to produce a given injury as heavy clay or silt soils.

13. The commercial ammonium phosphate was no more toxic than the pure mono-ammonium phosphate.

14. Liming decreased the growth of the young corn plants, at the lower rates of application of the fertilizers in the germination experiments, but favored the plants at the higher rates. Using various combinations of fertilizers did not lessen the injury of individual salts to any extent, if at all.

15. Corn, buckwheat, barley, wheat and oats were resistant to large applications of fertilizers, while vetch, rape and cowpeas were relatively susceptible.

16. Under laboratory conditions, using tumblers holding 200 gm. of soil, the injury produced by a given amount of the fertilizers was practically the same whether applied in direct contact with the seed or uniformly mixed with the soil.

17. Under field conditions applications of 100 pounds of ammonium phosphate per acre in the row did not injure the germination of corn, while the 150-pound applications showed a slight injury.

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REFERENCES

(1) Beseler, O., and Maercker, M. 1883 Versuche über den Einfluss der Aussaatstärke und der anwendung künstlicher Düngemittel auf den Ertrag und die Zusammensetzung des Hafers. *In* Centbl. Agr. Chem., Bd. 12, p. 472–480; Bd. 13 (1884), p. 453–461.

(2) Boussingault 1858 Sur l'action de salpetre employe comme engrais. *In* Jour. Agr. Prat., t. 2, p. 446–449.

(3) Claudel, and Cochetelle, J. 1896 Effect of some substances used as manures on germination. *In* Ann. Agron., t. 22, p. 131–142.

(4) Cook, R. C., and Allison, F. E. 1917 Effect of soil reaction upon the availability of ammonium sulfate. *In* Soil Sci., v. 3, p. 487–498.

(5) Ehrenberg, P. 1907 The movement of ammoniacal nitrogen in nature. *In* Mitt. Landw. Inst. Breslau, Bd. 4, Heft 1–2, p. 47–300.

(6) Hall, A. D., and Gimingham, C. T. 1907 The interaction of ammonium salts and the constituents of the soil. *In* Jour. Chem. Soc. [London], v. 91–92, no. 534, p. 677–687.

(7) Hals, S. 1902 Ammoniacal and nitric nitrogen. *In* Tidsskr. Norske Landbr., v. 9, no. 5, p. 212–225.

(8) Harris, F. S. 1915 Effect of alkali salts in soils on the germination and growth of crops. *In* Jour. Agr. Research, v. 5, no. 1, p. 1–53.

(9) Harris, F. S. 1915 The movement of soluble salts with the soil moisture. *In* Utah Agr. Exp. Sta. Bul. 139, p. 119–124.

(10) Immendorff, H. 1892 Beiträge zur Lösung der "Stickstoffrage." *In* Landw. Jahrb., Bd. 31, p. 281–339.

(11) Johnson, S. W. 1883 How Crops Feed, p. 301–304. Orange Judd Co., New York.

(12) Kearney, T. H., and Harter, L. L. 1907 Comparative tolerance of various plants for the salts common in alkali soils. U. S. Dept. Agr. Bur. Plant Indus. Bul. 113, p. 22.

(13) Kretschiner, et al. 1906 Field experiments on the fertilizing action of sulphate of ammonia and nitrate of soda. *In* Arb. Deut. Landw. Gesell., No. 121, 234 + xxiii p.

(14) LEMMERMANN, O., et al. 1911 Investigations on the behavior of ammoniacal nitrogen in limed and unlimed soil. *In* Landw. Jahrb., Bd. 40, p. 173–244, 255–324.

(15) LEMMERMANN O., et al. 1912 Increasing of ammonia-fixing power of soils under the influence of calcium carbonate. *In* Fuhling's Landw. Ztg., Bd. 61, No. 7, p. 240–253.

(16) LINT, H. C., AND COLEMAN, D. A. 1916 Sources of error in bacteriological analysis of soils. *In* Soil Sci., v. 2, no. 2, p. 157–163.

(17) LIPMAN, C. B., AND GERICKE, W. F. 1916 A vegetation experiment on the nitrogenous fertilizers in an arid soil. *In* Soil Sci., v. 2, p. 575–581.

(18) LIPMAN, J. G., AND BROWN, P. E. 1907 Ammonification in culture solutions as affected by soil treatment. *In* N. J. Agr. Exp. Sta. 28th Ann. Rpt. (1907), p. 195–199.

(19) LIPMAN, J. G., BROWN, P. E., AND OWEN, I. L. 1909 Comparative tests on the ammonification and nitrification of nitrogenous materials. *In* N. J. Agr. Exp. Sta. 30th Ann. Rpt. (1909), p. 169–174.

(20) LIPMAN, J. G., BROWN, P. E., AND OWEN, I. L. 1910 The relative availability of nitrogenous materials as measured by nitrification. *In* N. J. Agr. Exp. Sta. 31st Ann. Rpt. (1910), p. 151–155.

(21) LIPMAN, J. G., BLAIR, A. W., OWEN, I. L., AND MCLEAN, H. C. 1912 Report of the soil chemist and bacteriologist. *In* N. J. Agr Exp. Sta. 33rd Ann. Rpt. (1912), p. 209.

(22) LIPMAN, J. G., AND BLAIR, A. W. 1916 Investigations relative to the use of nitrogenous plant-foods, 1898–1912. N. J. Agr. Exp. Sta. Bul. 288, 124 p.

(23) MALPEAUX, L., AND LEFORT, G. 1912 The circulation of nitrates in the soil. *In* Ann. Sci. Agron., s. 4, ann. 1, t. 2, p. 241–258.

(24) MAYER, A. 1901 Ueber die Bedingungen des Entstehens der Eiweissstoffe in der Pflanze. *In* Landw. Vers. Stat., Bd. 55, p. 453–461.

(25) MÜNTZ, A., AND GAUDECHON, H. 1908 The diffusion of fertilizer salts in the soil. *In* Ann. Inst. Nat. Agron., s. 2, t. 7, no. 2, p. 205–238.

(26) MÜNTZ, A., AND NOTTIN, P. 1907 Studies on the value of calcium cyanamid as a nitrogenous fertilizer. *In* Ann. Inst. Nat. Agron., s. 2, t. 6, no. 1, p. 145–183.

(27) PANTANELLI, E., AND SEVERINI, G. 1910 Some experiments on the utilization of ammonium salts by green plants. *In* Staz. Sper. Agr. Ital., v. 43, no. 6, p. 449–544.

(28) RITTHAUSEN, H., AND POTT, R. 1873 Untersuchungen über den Einflus einer an Stickstoff und Phosphorsäure de Pflanze und der Samen von Sommerweizen. *In* Landw. Vers. Stat. Bd. 16, p. 384.

(29) RUSCHE, A. 1912 Influence of various fertilizers on the germination of field crop seeds. *In* Jour. Landw., Bd. 60, No. 4, p. 305–365.

(30) SCHÄFER-HEIDE, A. 1906 Die Ausnutzung des Ammoniakstickstoff. *In* Compt. Rend. Acad. Sci. [Paris], t. 140, p. 1190.

(31) SCHREIBER, C. 1908 Recent investigations of the nitrogenous fertilizers. *In* Rev. Gén. Agron., n. s., t. 3, no. 11, 433–440; no. 12, p. 481–486; t. 4, no. 1, p. 4–9.

(32) STEFFECK, H., AND MAERCKER, M. 1896 Efficacy of various manures. *In* Jahrb. Agr. Chem. Vers. Stat. Halle, 1896, p. 105–131.

(33) TANRET, CHAS. 1897 Action of ammonium salts on *Aspergillus niger*. *In* Bul. Soc. Chim. Paris, s. 3, t. 17, p. 914–921.

(34) WAGNER, P. 1892 Die Stickstoffdüngung der Landwirtschaftlichen Kulturpflanzen, Berlin, 1892. *Abs. in* Centbl. Agr. Chem., Bd. 21, p. 791–792.

(35) WAGNER, P., HAMANN, G., AND MÜNZINGER, A. 1907 Experiences aver le nitrate de soundre, les sels diamamonium et la chaux azote comme engrais. *In* Arb. Deut. Landw. Gesell., 1907, No. 129, p. vi + 286.

(36) WILFARTH, H., RÖMER H., AND WIMMER, G. 1906 Über die Nahrstoffaufnahme der Pflanzen in verschiedenen Zeiten ihres Wachstums. *In* Landw. Vers. Stat., Bd. 63, p. 1–70.

THE SIGNIFICANCE OF THE SULFUR IN SULFATE OF AMMONIA APPLIED TO CERTAIN SOILS

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The superiority of sulfate of ammonia to nitrate of soda, nitrate of lime, and other nitrogenous fertilizers for the production of barley, in certain soil types in California, which we have demonstrated (1) led us to inquire into the more direct nature of its cause. While we had shown the high coefficient of nitrifiability which characterizes sulfate of ammonia in those soils to be the cause of its superiority over other nitrifiable materials, it did not appear entirely clear why nitrate of soda and particularly nitrate of calcium should not be equally good with moisture conditions being maintained at the optimum. After studying the situation, it occurred to us that the sulfur of the ammonium sulfate might be the cause of its superiority over the nitrate fertilizers. This seemed particularly reasonable since the soils under study are markedly deficient in organic matter and hence do not support a vigorous sulfofying flora. We, therefore, inaugurated an experiment to determine the rôle of sulfur, in barley growth on one type of the soils in question, when used in conjunction with nitrogenous fertilizers. It was argued that if sulfur in one or more forms, when combined with nitrates, induced similar yields to those obtained with sulfate of ammonia, the sulfur must be the cause of the superiority of the ammonium salt over the nitrates in the cases in question.

DESCRIPTION OF THE EXPERIMENT

The soil employed was the Oakley blow sand, which we have already described in detail elsewhere (2). It was placed in glazed earthenware jars such as those commonly used in greenhouse work. Every test was made in triplicate jars, except in some cases in which the lack of sufficient soil rendered it necessary to employ only duplicate jars. Two parallel series were run. In the first, the various nitrogenous fertilizers were used as in the experiments above cited. In the second, the same nitrogenous fertilizers were used in the same way, but sulfur in various forms was added to each. The amount of nitrogen employed was equivalent in all cases to that added on the basis of a field application of dried blood at the rate of 1000 pounds per acre. Similarly, sulfur was used in the same quantity for all cultures to which it was applied, regardless of the form used and was equivalent to the amount of sulfur contained in the sulfate of ammonia application alone. Nitrogen was

employed in the forms of sulfate of ammonia, nitrate of soda, nitrate of lime, and dried blood. Sulfur was used in the forms of flowers of sulfur, sulfuric acid, and sodium sulfate. The applications of the various materials were made in accordance with the usual procedure which is based on the properties of every one of the substances. The moisture content of the soil was maintained uniformly in all the jars and at as near the optimum point as possible. Twelve barley seeds of the Beldi variety, from selected heads, were planted in every jar as in all other experiments, and the plants were thinned to six after they had attained a height of 3 to 4 inches. At the end of the growing season, determinations were made of the number of heads of grain per jar, the number of tillers, the total height of tillers, and the dry weights of straw, of grain, and of roots. Control jars were used, for growing plants, in which no fertilizer and no sulfur were employed and others in which only sulfur, but no nitrogen was applied. The data gathered are presented in the accompanying tables. It is at once apparent even from a brief study of the data given, that, in the first place, sulfate of ammonia is far superior to the other nitrogenous fertilizers for barley growth under the conditions described above. Secondly, it is clear that sulfur in all forms, when supplementing the nitrate or blood, induces marked increases in barley growth over and above that induced by the nitrogen alone. Thirdly, the sulfur without the nitrogen is practically without effect on barley production on the Oakley blow sand.

Considering the figures more in detail, it is obvious in the first table that marked effects, for the better, result from the application of nitrogen as regards the height of the stalks produced. The number of stalks or of heads produced, however, is only slightly affected, if at all, by all forms of nitrogen except sulfate of ammonia, which stimulates their production very markedly. When however, sulfur is added to the nitrogen, very striking increases in the number of heads and stalks occur in the case of all the jars, without any marked increase in the total height of the stalks. Moreover, the increases noted are, in most cases, sufficient to render the total growth about equivalent in those jars to that obtained in the sulfate of ammonia jars. This is true when any form of sulfur is added to nitrate of soda, but flowers of sulfur are not as good in that direction as sulfuric acid or sodium sulfate. On the other hand, it is true for nitrate of lime only in the cases of flowers of sulfur and sodium sulfate, but not in that of sulfuric acid, which actually depresses growth below that obtained when only nitrate of lime is applied. In the case of dried blood, sodium sulfate was not used and of the other two forms of sulfur, one—sulfuric acid—stimulated the plants in the directions indicated markedly, while the other—flowers of sulfur—was almost without effect.

Coming now to the more accurately measurable criteria for judging of the effects of nitrogen and sulfur on the barley plants, viz., the weights of straw, grain, and roots, the following observations are easily made. The application of sulfate of ammonia to the Oakley sand resulted in the production of about fourteen times as much total dry matter as that produced in the untreated

control jars. The jars treated with nitrate of soda, nitrate of lime, or dried blood, produced only a little more than half as much dry matter as those treated with sulfate of ammonia, the blood being slightly superior to the other two. When any form of sulfur is added to the Oakley sand without nitrogen,

TABLE 1

Number and height of shoots and number of heads

NUMBER	NITROGENOUS FERTILIZER	FORM OF SULFUR	NUMBER OF PLANTS PER POT	NUMBER OF HEADS PER POT	NUMBER OF SHOOTS PER POT	TOTAL HEIGHT OF SHOOTS	AVERAGE HEIGHT OF SHOOTS
						inches	*inches*
1	$(NH_4)_2SO_4$		6	14	15	475.5	31.7
2	$(NH_4)_2SO_4$		6	12	12	384.5	32.0
3	$(NH_4)_2SO_4$		6	12	12	404.0	33.7
4	$NaNO_3$		6	6	6	218.5	36.4
5	$NaNO_3$		6	8	8	253.0	31.6
6	$NaNO_3$		6	6	6	215.5	35.9
7	$Ca(NO_3)_2$		6	6	6	207.0	34.5
8	$Ca(NO_3)_2$		6	7	8	246.0	30.8
9							
10	Blood		6	8	9	254.5	28.3
11	Blood		6	7	8	247.0	30.9
12		Na_2SO_4	6	6	6	106.0	17.7
13		Na_2SO_4	6	6	6	92.0	15.3
14		H_2SO_4	6	6	6	104.0	17.3
15		H_2SO_4	6	6	6	100.0	16.6
16		Sulfur	6	6	6	99.0	16.5
17		Sulfur	6	6	6	124.0	20.7
18	$NaNO_3$	Na_2SO_4	6	12	13	414.0	31.8
19	$NaNO_3$	Na_2SO_4	6	10	10	312.5	31.2
20	$NaNO_3$	H_2SO_4	6	12	12	403.0	33.6
21	$NaNO_3$	H_2SO_4	6	10	13	434.5	33.4
22	$NaNO_3$	Sulfur	6	10	11	315.0	28.6
23	$NaNO_3$	Sulfur	6	9	9	305.0	33.9
24	$Ca(NO_3)_2$	Na_2SO_4	6	14	15	412.0	27.5
25	$Ca(NO_3)_2$	Na_2SO_4	6	10	10	323.0	32.3
26	$Ca(NO_3)_2$	H_2SO_4	6	6	6	182.0	30.3
27	$Ca(NO_3)_2$	H_2SO_4	6	6	6	203.0	37.1
28	$Ca(NO_3)_2$	Sulfur	6	12	12	388.5	32.3
29	$Ca(NO_3)_2$	Sulfur					
30	Blood	H_2SO_4	6	11	11	327.0	29.7
31	Blood	H_2SO_4	6	12	12	382.0	31.8
32	Blood	Sulfur	6	9	9	276.0	30.7
33	Blood	Sulfur	6	7	9	286.0	31.8
34	Control		6	6	6	95.0	15.9
35	Control		6	6	6	110.0	18.3

it is without effect as regards the dry matter produced, just as it was described above to be, in so far as the numbers of heads and stalks are concerned. When, however, sulfur in any form is added, as described above, to the nitrates or to dried blood, it induces marked increases in the production of dry matter over

that produced by the nitrogen alone and, in most cases, the quantity of such increase is great enough to bring the total yield up to that of the sulfate of ammonia jars or nearly so. As was true in the case of the number of heads and stalks produced, flowers of sulfur appears to be less efficient than the other

TABLE 2

Yields of straw, grain and roots obtained in experiment

NUMBER	NITROGENOUS FERTILIZER	FORM OF SULFUR	WEIGHT OF STRAW	AVERAGE WEIGHT OF STRAW	WEIGHT OF GRAIN	AVERAGE WEIGHT OF GRAIN	WEIGHT OF ROOTS	AVERAGE WEIGHT OF ROOTS	TOTAL DRY WEIGHT	AVERAGE TOTAL DRY WEIGHT
			grams	*grams*	*grams*	*grams*	*grams*	*grams*	*grams*	*grams*
1	$(NH_4)_2SO_4$		23.94	23.38	16.06	15.69	5.27	6.84	45.27	45.90
2	$(NH_4)_2SO_4$		23.17		14.83		7.20		45.20	
3	$(NH_4)_2SO_4$		23.02		16.18		8.04		47.24	
4	$NaNO_3$		11.65	11.99	8.15	8.95	5.60	4.91	25.40	25.85
5	$NaNO_3$		14.05		10.95		4.95		29.95	
6	$NaNO_3$		10.26		7.74		4.20		22.20	
7	$Ca(NO_3)_2$		11.18	13.09	6.82	8.91	5.40	6.60	23.40	28.60
8	$Ca(NO_3)_2$		15.00		11.00		7.80		33.80	
9	Lost									
10	Blood		14.46	13.97	11.03	11.28	5.06	5.08	30.55	30.33
11	Blood		13.47		11.53		5.10		30.10	
12		Na_2SO_4	1.24	1.35	1.46	1.40	0.40	0.40	3.10	3.15
13		Na_2SO_4	1.46		1.34		0.40	0.40	3.20	
14		H_2SO_4	0.94	1.52	1.46	1.18	0.22	0.37	2.62	3.07
15		H_2SO_4	2.10		0.90		0.52		3.52	
16		Sulfur	1.54	1.97	1.46	2.53	0.60	1.25	3.60	5.75
17		Sulfur	2.40		3.60		1.90		7.90	
18	$NaNO_3$	Na_2SO_4	20.94	20.66	15.06	14.84	8.26	8.73	44.26	44.23
19	$NaNO_3$	Na_2SO_4	20.38		14.62		9.20		44.20	
20	$NaNO_3$	H_2SO_4	22.00	23.90	16.00	16.40	7.70	6.60	45.70	46.90
21	$NaNO_3$	H_2SO_4	25.80		16.79		5.50		48.09	
22	$NaNO_3$	Sulfur	18.17	17.32	12.43	12.30	5.00	6.30	35.60	35.92
23	$NaNO_3$	Sulfur	16.47		12.17		7.60		36.24	
24	$Ca(NO_3)_2$	Na_2SO_4	23.35	26.61	16.65	15.39	9.50	7.75	49.50	44.75
25	$Ca(NO_3)_2$	Na_2SO_4	29.87		14.13		6.00		40.00	
26	$Ca(NO_3)_2$	H_2SO_4	7.50	9.56	5.60	6.69	2.15	3.38	15.25	19.63
27	$Ca(NO_3)_2$	H_2SO_4	11.62		7.78		4.60		24.00	
28	$Ca(NO_3)_2$	Sulfur	18.80	18.80	14.20	14.20	1.08	10.80	43.80	43.80
29	$CaNO_3$	Sulfur	Contaminated by rain							
30	Blood	H_2SO_4	17.55	18.08	13.64	14.22	11.00	9.40	42.19	41.70
31	Blood	H_2SO_4	18.60		14.80		7.80		41.20	
32	Blood	Sulfur	17.62	17.26	13.08	12.09	7.80	9.40	38.50	38.75
33	Blood	Sulfur	16.90		11.10		11.00		39.00	
34	Control		1.54	1.64	1.26	1.26	0.40	0.45	3.20	3.75
35	Control		1.74		1.26		0.50		3.50	

forms in increasing dry-matter production over that of the nitrogen-treated pots alone, but is none the less markedly stimulating in that respect. It seems, moreover, to be slightly more efficient when added to dried blood than when

added to nitrate of soda but the difference is not great. On the other hand, sulfuric acid is more efficient when supplementing nitrate of soda than when supplementing dried blood. As regards total dry-matter production, sulfuric acid acts similarly when added to calcium nitrate as it does with regard to the production of heads and stalks.

Sodium sulfate is equally stimulating when used with nitrate of soda or with nitrate of lime, and sulfuric acid is likewise so for nitrate of soda and for dried blood. Flowers of sulfur show similarity to the other forms of sulfur in maximum stimulation with regard to one case only, viz., when it is added to nitrate of lime. Unfortunately, one of the duplicate jars in this case was lost and the figure obtained is, therefore, not as reliable as it would otherwise have been.

In general, our data seem to show beyond question that, barring certain pronounced side-effects, such as that evidently obtaining in the nitrate of lime-sulfuric acid cultures, equally good results in barley production can be obtained on soils of the type in question with the nitrates and with blood as with sulfate of ammonia, if sulfur is added in proper form and quantity. Just why sulfuric acid, when added to nitrate of lime, should produce an effect so strikingly different from that produced by it when added to nitrate of soda seems at first sight not very clear. A little reflection brings forward the thought, however, that the greater solubility of the sodium sulfate than that of the calcium sulfate, the salts resulting in the two cases, may account for the supply of sufficient available sulfur in one case and not in the other; and hence the difference in effects on the barley plant. This explanation is not entirely satisfying, however, owing to the very marked depression in the growth of the barley plants induced by sulfuric acid when it is added to calcium nitrate. Another factor entering into the problem may be the loss of nitric acid which may be greater in one case than the other when the acid is added to the nitrate.

The fact above noted of the non-effectiveness of all the forms of sulfur when applied to the soil without nitrogen, requires perhaps another word of comment. If, as seems true from our data, sulfur is needed by the barley plant on the soil in question, why should not sulfur exert at least a part of its beneficial effect even when nitrogen is not added with it? The answer may be in the theoretical consideration, which we propose, to the effect that nitrogen, being needed in this case far more than sulfur, the latter is powerless to influence the plant in the building of further protein substances, to the building of which that nitrogen is absolutely essential.

In view of the striking nature of the results above described, the method of improving the Oakley blow sand and similar soils covering large areas in this state is clearly indicated. Nitrogen in some readily available form must be used as fertilizer until such time as it shall be possible to increase the nitrogen and organic matter content of the soil by means like green manuring, and the efficient use of nitrogen-fixing bacteria. The choice of the most profitable form of nitrogen can be determined by reference to the tables

submitted herewith, taken together with the prevailing prices of the materials in question. For the best results with annual crops, to which our remarks particularly apply, sulfur in some form, probably as flowers of sulfur, will have to be employed to supplement the nitrogenous fertilizer unless sulfate of ammonia is used.

No mention is made here of the work of investigators in this country and in Europe which have testified to the value of sulfur as a soil amendment because they have no direct bearing on the subject of this paper.

SUMMARY

1. The superiority of sulfate of ammonia over other readily available nitrogenous fertilizers for barley on the Oakley blow sand as demonstrated in earlier experiments is confirmed in a new series of tests.

2. The cause of such superiority of sulfate of ammonia was sought for and believed to be found in its sulfur content.

3. A statement of the mode of procedure employed in the experiment, and also details of the results obtained and their discussion are submitted above.

REFERENCES

(1) LIPMAN, C. B., AND GERICKE, W. F. 1916 A vegetation experiment on the availability of nitrogenous fertilizers in an arid soil. *In* Soil Sci, v. 2, no. 6, p. 575–581.

(2) LIPMAN, C. B., AND GERICKE, W. F. 1917 Experiments on the effects of constituents of solid smelter wastes on barley growth in pot cultures. *In* Univ. Cal. Pub. Agr. Sci., v. 1, no. 13, p. 495–587.

TOXICITY OF MONOBASIC PHOSPHATES TOWARDS SOYBEANS GROWN IN SOIL- AND SOLUTION-CULTURES

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The mineral elements essential to the growth of plants in the soil or in culture media, may under certain conditions become extremely toxic to the plants. In a nutritive medium with a given total concentration of the necessary salt constituents for plants, marked changes in growth are sure to occur with any considerable alteration in the salt proportions. The salt proportions may be altered in such a manner as to produce considerable change in the growth rates without causing any specific injury to the plants. When, on the other hand, the physiological balance of the medium is too greatly disturbed by the alteration of the salt proportions, toxic effects in some specific form are quite sure to appear, and these can usually be related to one or more of the constituents of the medium. Thus, in a series of 84 four-salt nutritive solutions, all having the same total concentration but with varying proportions of the same four salts, Tottingham (14) found that a number of his solutions caused specific injury to wheat seedlings, while other solutions of the same series produced excellent growth without toxic effects. Tottingham was able to relate the injury to the magnesium content of the solutions. He pointed out, however, that the conditions determining the degree of the injury were related to the relative proportions of all the constituents rather than to the partial concentration of magnesium alone. In a more recent investigation (12) of the same character, with solutions comprising the three salts, mono-potassium phosphate, calcium nitrate, and magnesium sulfate, in 36 different proportions but with constant total concentrations, it was found that out of the total of 36 solutions, 7 caused very severe injury to wheat seedlings and 14 produced slight injury. The remaining 15 solutions showed various degrees of productiveness without toxic effects. The disturbance here observed was practically identical with that described by Tottingham (14) and called by him magnesium injury.

In a nutritive medium with constant relative proportions of the necessary salt constituents for plants, variation in the growth rates without toxic effects may be brought about also by alterations in the total concentration of the medium. This is primarily an osmotic relation and appears to be dependent upon the ease with which water may be taken into the cells. Trelease (15) has shown that the growth rate (as indicated by dry-weight yields) is an ap-

proximate linear relation to the concentration of the medium above the optimum, the growth rate decreasing with an increase in concentration. If, however, the total concentration of the medium is gradually increased without alteration of the salt proportions, a point is finally reached where the plants may show symptoms of specific poisoning due to the absorption of excessive amounts of one or more of the essential constituents. This has been shown to occur with total concentrations not above the optimum for plant growth. Tottingham's series of 4-salt solutions with a total osmotic concentration value of 0.5 atmospheres produced no magnesium injury to wheat seedlings. When the concentration of the solutions was increased to an osmotic value of 2.5 atmospheres and 8.15 atmospheres, many of the solutions produced severe injury. The solutions of the 3-salt series employed by the present writer (12) caused no toxic symptoms in wheat seedlings when the total osmotic concentration value was 0.10 atmospheres, but with the same relative salt proportions at total concentrations having osmotic values of 1.75 atmospheres and 4 atmospheres, many of the solutions produced severe injury.

In the course of some experimental work (13) concerning the influence of various salts on the growth of soybeans in soil, it was observed that several phosphate salts, when added singly to the soil, even in comparatively small applications, proved to be toxic to the plants, which not only were retarded in their development, but also suffered specific injury. It was the purpose of the present investigation to study somewhat in detail the nature of this injury and to determine the general effects of several monobasic phosphate salts on the growth of soybeans under various experimental conditions. These tests involved three distinct sets of experiments employing five different monobasic phosphate salts: (a) in soil to which the salts were added singly in the form of solutions with varying concentrations; (b) in connection with a complete fertilizer ration added to the soil in mixed solutions with constant total concentrations; and (c) in culture solutions with complete nutrient mixtures.

EXPERIMENTAL METHODS

1. Preparation of stock solutions

Throughout these tests Baker's analyzed chemicals were used, including mono-sodium phosphate, mono-ammonium phosphate, mono-potassium phosphate, mono-calcium phosphate, mono-magnesium phosphate, and magnesium sulfate in the crystalline form, and calcium nitrate in the form of fused lumps (labeled as containing $4H_2O$). In all the work with soil cultures, the salts were introduced into the soil in the form of solutions. Stock solutions of the salts, except mono-calcium phosphate, were prepared by dissolving separately in volumetric flasks a gram molecular weight of each salt, after which the solutions were made up to liter volume at 15°C. The distilled water used throughout was obtained from a "Barnstead" still and was stored in large glass bottles, from which it was drawn for immediate use. The stock solution of mono-cal-

cium phosphate was prepared by dissolving one-eighth (0.125m) gram molecular weight of the salt in a volumetric flask, keeping the mixture at a temperature below 15°C. until the process of solution was completed, in order to avoid decomposition and precipitation, which sometimes occurred when the solutions were prepared at room temperature. The flask was then filled to liter volume at 15°C.

For immediate use, the volume-molecular stock solutions were diluted to concentrations better suited to the preparation of the culture solutions. The concentration of the diluted stock solutions was one-fourth (0.25m) molecular, excepting the mono-calcium phosphate, which had a concentration of one-eighth (0.125m) molecular.

Throughout this work all glass vessels employed as containers of solutions or distilled water were thoroughly washed with bichromate-sulfuric acid cleansing mixture, and were then rinsed with tap water, followed by distilled water.

2. *Preparation of culture solutions*

a. Pure solutions. Throughout this study concentrations will be stated in terms of gram molecules per liter of solution (volume-molecular concentration) or in terms of possible osmotic pressure in atmospheres (osmotic concentration).

Five series of pure solutions corresponding to the five phosphate salts, were employed in soil cultures. Each series comprised 10 solutions. Eight solutions of each series, beginning with the first, varied in osmotic concentration from 0.5 atmospheres to 4 atmospheres, by increments of 0.5 atmospheres. The remaining two solutions of each series had concentrations of 5 and 7 atmospheres, respectively. Each series possessed, therefore, a range in concentration from an osmotic value of 0.5 atmospheres to an osmotic value of 7 atmospheres.

In table 1 are given the approximate osmotic concentration values in atmospheres, and the volume-molecular concentrations of the ten solutions of each series, as well as the per cent of total salt added to each soil culture, calculated on the basis of the weight of air-dry soil, which in all cases was 4 kgm. to each culture.

The osmotic concentration values of these solutions were calculated approximately according to the method employed by Tottingham (14) who based his calculations on the data of electrolytic dissociation as given by Jones (7). Electrolytic dissociation data for the phosphates here employed are not found in Jones' tables. The data required for the calculation of the osmotic concentration values of mono-potassium phosphate are given by Tottingham (14, p. 177), and similar data for mono-sodium and mono-ammonium phosphates are given by Watkins and Jones (16). Data for mono-calcium and mono-magnesium phosphates were not available. The osmotic concentrations of these salts in solutions were determined approximately by the method of cryoscopy (11).

b. Mixed solutions. Five series of mixed solutions were employed. Each of these consisted of the Tottingham (14) series of nutrient solutions in which a monobasic phosphate was substituted for the potassium nitrate. The series will be designated series I, II, III, IV, and V, and will correspond to the salts mono-sodium phosphate, mono-ammonium phosphate, mono-potassium phosphate, mono-calcium phosphate, and mono-magnesium phosphate, respectively. Thus each solution of a series contained, in addition to the three salts, mono-potassium phosphate, calcium nitrate, and magnesium sulfate, one of the five monobasic salts. The three salts, mono-potassium phosphate, calcium nitrate and magnesium sulfate, containing all the so-called essential constituents (excepting iron) of a complete nutrient mixture, were present in

TABLE 1

Approximate osmotic and volume-molecular concentrations of solutions of monobasic phosphates employed singly in soil cultures; also amount of salts in each culture expressed as per cent of the weight of air-dry soil

SOLUTION NUMBER	OSMOTIC CONCENTRATION	VOLUME-MOLECULAR CONCENTRATIONS					WEIGHT OF SALT AS PER CENT OF AIR-DRY SOIL				
		Series I NaH_2PO_4	Series II $(NH_4)H_2PO_4$	Series III KH_2PO_4	Series IV $CaH_4(PO_4)_2$	Series V $MgH_4(PO_4)_2$	Series I NaH_2PO_4	Series II $(NH_4)H_2PO_4$	Series III KH_2PO_4	Series IV $CaH_4(PO_4)_2$	Series V $MgH_4(PO_4)_2$
	atm.										
1	0.5	0.0100	0.0101	0.0104	0.0071	0.0076	0.015	0.015	0.018	0.021	0.021
2	1.0	0.0201	0.0204	0.0210	0.0143	0.0153	0.030	0.030	0.036	0.042	0.042
3	1.5	0.0305	0.0308	0.0321	0.0216	0.0231	0.046	0.044	0.055	0.063	0.063
4	2.0	0.0425	0.0426	0.0442	0.0286	0.0306	0.064	0.063	0.075	0.084	0.084
5	2.5	0.0539	0.0539	0.0560	0.0359	0.0384	0.081	0.077	0.095	0.105	0.105
6	3.0	0.0658	0.0658	0.0685	0.0431	0.0461	0.099	0.095	0.116	0.126	0.125
7	3.5	0.0769	0.0769	0.0800	0.0504	0.0540	0.115	0.111	0.136	0.148	0.147
8	4.0	0.0901	0.0900	0.0936	0.0578	0.0618	0.135	0.129	0.159	0.169	0.168
9	5.0	0.1132	0.1130	0.1176	0.0805	0.0860	0.170	0.162	0.199	0.236	0.234
10	7.0	0.1622	0.1615	0.1685	0.1051	0.1120	0.243	0.232	0.281	0.307	0.306

corresponding solutions of the five series in the same osmotic and volume-molecular proportions. The different solutions all had approximately the same total osmotic concentration (2.5 atmospheres), but each solution of the same series differed from all the others in the relative proportions of the four component salts.

In addition to these five series, a sixth series was employed as a control series; this, for the sake of convenience, may be called the 3-salt series. In the preparation of the 3-salt series, mono-potassium phosphate, calcium nitrate, and magnesium sulfate only were employed. These three salts were present in each solution of this series in the same relative proportions as were the same three salts in corresponding solutions of the other five series. However, the total osmotic concentrations of the solutions of the 3-salt series were

made equal to those of the solutions of the other five series (2.5 atmospheres), each containing four constituent salts. Thus the partial volume-molecular concentrations of the three salts in each solution of the 3-salt series differed from the partial volume-molecular concentrations of the same salts in corresponding solutions of the other series, but the relative proportions of these three salts, in corresponding solutions of the different series, remained the same.

The method here adopted to control the osmotic concentrations was the same as that employed by Tottingham (14, p. 192). The relative proportions of the salts were varied in such a way that a decrease in the partial concentration of one salt was balanced by a sufficient increase in the partial concentrations of the remaining salts to keep the total osmotic concentration of the solution constant. The total osmotic concentration (2.5 atmospheres) for each 4-salt series was considered to be divided into ten equal parts and these parts were distributed among the four salts in all possible proportions. Thus each salt produced in the different solutions of a series, from one-tenth to seven-tenths of the total osmotic concentration.

A detailed discussion of the methods by which the osmotic concentration of a series of solutions, differing in the proportions of the constituent salts, may be controlled, as well as a discussion of the methods of calculation by which the partial osmotic concentrations may be approximately determined for complex mixtures with constant total osmotic concentrations, has been presented by Tottingham (14, p. 177–182, 192).

Variations in the proportions of the four salts, as here employed, by increments of one-tenth of the total osmotic concentration yield a series of 84 solutions. Six such series, therefore, comprise 504 solutions. It was found impracticable to conduct so large a number of cultures during the same time period. In the present work, therefore, the number of solutions employed was reduced to 20 in each series. These were selected to correspond to 20 representative solutions from the Tottingham (14) series of 84 solutions As is well known, the scheme adopted by Tottingham to represent diagrammatically the relative composition of the solutions of his 4-salt series, makes use of six triangles (T_1, T_2, T_3, etc.) and a point (T_7). The rows of each triangle are numbered from the base to the apex (R_1, R_2, R_3 etc.) and the solutions or cultures in each row are numbered from left to right (C_1, C_2, C_3, etc.), A detailed description of this arrangement is presented by Tottingham (14, p. 192–195) and need not be repeated here. The twenty cultures of the Tottingham series corresponding to those of each series here employed, were selected in the following manner. From the Tottingham diagrammatic scheme the three triangles T_2, T_4, and T_6 were omitted, leaving the three triangles T_1, T_3, and T_5 and the point T_7. By omitting from these remaining three triangles every other row, starting with the second row from the base of each triangle, and by further omitting every other solution from the remaining rows, starting with the second solution in each row, a total of twenty, out of

the entire series of 84 solutions, remain. These twenty solutions are uniformly distributed throughout the series.

The first column of table 2 presents the solution numbers as they occur in the Tottingham series. It will be observed that the triangles, rows, and cultures represented by even numbers are omitted while all those represented by odd numbers are retained and appear in the table. The table gives the partial volume-molecular concentration of each solution of the five series. In the last three columns are given the partial volume-molecular concentra-

TABLE 2

Partial volume-molecular concentrations of five monobasic phosphates each employed with KH_2PO_4 $Ca(NO_3)_2$ *and* $MgSO_4$ *to form five series of culture solutions with total osmotic concentrations of 2.5 atmospheres*

SOLUTION NUMBER	PARTIAL CONCENTRATIONS, VOLUME-MOLECULAR							
	Series I NaH_2PO_4	Series II $(NH_4)H_2PO$	Series III KH_2PO_4	Series IV $CaH_4(PO_4)_2$	Series V $MgH_4(PO_4)_2$	Component salts of each solution, series I to V		
						KH_2PO_4	$Ca(NO_3)_2$	$MgSO_4$
$T_1R_1C_1$	0.0050	0.0051	0.0052	0.0036	0.0038	0.0052	0.0036	0.0406
C_3	0.0050	0.0051	0.0052	0.0036	0.0038	0.0052	0.0108	0.0290
C_5	0.0050	0.0051	0.0052	0.0036	0.0038	0.0052	0.0180	0.0174
C_7	0.0050	0.0051	0.0052	0.0036	0.0038	0.0052	0.0252	0.0058
R_3C_1	0.0050	0.0051	0.0052	0.0036	0.0038	0.0156	0.0036	0.0290
C_3	0.0050	0.0051	0.0052	0.0036	0.0038	0.0156	0.0108	0.0174
C_5	0.0050	0.0051	0.0052	0.0036	0.0038	0.0156	0.0180	0.0058
R_5C_1	0.0050	0.0051	0.0052	0.0036	0.0038	0.0260	0.0036	0.0174
C_3	0.0050	0.0051	0.0052	0.0036	0.0038	0.0260	0.0108	0.0058
R_7C_1	0.0050	0.0051	0.0052	0.0036	0.0038	0.0364	0.0036	0.0058
$T_3R_1C_2$	0.0150	0.0153	0.0156	0.0108	0.0114	0.0052	0.0036	0.0290
C_3	0.0150	0.0153	0.0156	0.0108	0.0114	0.0052	0.0108	0.0174
C_5	0.0150	0.0153	0.0156	0.0108	0.0114	0.0052	0.0180	0.0058
R_3C_1	0.0150	0.0153	0.0156	0.0108	0.0114	0.0156	0.0036	0.0174
C_3	0.0150	0.0153	0.0156	0.0108	0.0114	0.0156	0.0108	0.0058
R_5C_1	0.0150	0.0153	0.0156	0.0108	0.0114	0.0260	0.0036	0.0058
$T_5R_1C_1$	0.0250	0.0255	0.0260	0.0180	0.0190	0.0052	0.0036	0.0174
C_3	0.0250	0.0255	0.0260	0.0180	0.0190	0.0052	0.0108	0.0058
R_3C_1	0.0250	0.0255	0.0260	0.0180	0.0190	0.0156	0.0036	0.0058
$T_7R_1C_1$	0.0350	0.0357	0.0364	0.0252	0.0266	0.0052	0.0036	0.0058

tions of the three salts KH_2PO_4, $Ca(NO_3)_2$ and $MgSO_4$, as they were employed with each of the five phosphate salts to form the five different series. Thus, the four salts employed in series I were present in the various solutions in the partial volume-molecular concentrations as given in the second column for NaH_2PO_4, and in the last three columns for KH_2PO_4, $Ca(NO_3)_2$ and $MgSO_4$, respectively. In the same manner are given successively the partial volume-molecular concentrations of the solutions of series II, III, IV and V.

It will be observed that the solutions of series III contain only the three salts, KH_2PO_4, $Ca(NO_3)_2$ and $MgSO_4$, but the partial osmotic concentration due to the KH_2PO_4 in each solution of this series is the same as that supplied by the total phosphates in the corresponding solutions of the other four series. The solutions of series III, therefore, will be treated throughout the study as though they were 4-salt solutions.

Table 3 gives the partial volume-molecular concentrations of the solutions of the 3-salt series. It is to be noted that while the relative proportions of the salts in each solution represented in this table are the same as are those of

TABLE 3

Partial volume-molecular concentrations of KH_2PO_4, $Ca(NO_3)_2$, *and* $MgSO_4$ *in culture solutions having total osmotic concentration of 2.5 atmospheres (3-salt series)*

SOLUTION NUMBER	PARTIAL CONCENTRATIONS, VOLUME-MOLECULAR		
	KH_2PO_4	$Ca(NO_3)_2$	$MgSO_4$
$T_1R_1C_1$	0.0058	0.0040	0.0451
C_3	0.0058	0.0120	0.0322
C_5	0.0058	0.0200	0.0193
C_7	0.0058	0.0280	0.0064
R_3C_1	0.0173	0.0040	0.0322
C_3	0.0173	0.0120	0.0193
C_5	0.0173	0.0200	0.0064
R_5C_1	0.0289	0.0400	0.0193
C_3	0.0289	0.0120	0.0064
R_7C_1	0.0404	0.0040	0.0064
$T_3R_1C_1$	0.0074	0.0051	0.0414
C_3	0.0074	0.0154	0.0248
C_5	0.0074	0.0257	0.0083
R_3C_1	0.0223	0.0051	0.0248
C_3	0.0223	0.0154	0.0083
R_5C_1	0.0371	0.0051	0.0083
$T_5R_1C_1$	0.0104	0.0072	0.0348
C_3	0.0104	0.0216	0.0116
R_3C_1	0.0312	0.0072	0.0116
$T_7R_1C_1$	0.0173	0.0120	0.0193

the same three salts [KH_2PO_4, $Ca(NO_3)_2$ and $MgSO_4$] in the corresponding solutions of table 2, the partial volume-molecular and partial osmotic concentrations are not the same, since the total osmotic concentration (2.5 atm.), which is the same for the solution of all the series, is made up of three salts in the solutions of this one series, and of four salts in the solutions of each of the other five series, except series III.

In preparing the individual culture solutions, the required amount of stock solution of the phosphate salt to be employed was drawn from a burette into a 500-cc. volumetric flask partially filled with water. To this was added in a

similar manner the required amounts of the stock solution of mono-potassium phosphate, calcium nitrate, and magnesium sulfate, in the order given. The flask was then filled to volume with distilled water.

3. The soil cultures

The soil employed throughout this work consisted of a mixture of white quartz sand and a rich sandy loam. The latter was obtained from the experiment grounds of the botanical department of the New Jersey Agricultural Experiment Station. The soil and sand were air-dried separately, after which they were sifted through a wire screen of 2-mm. mesh, and a sufficient quantity of each was stored for use during the work. Equal parts by volume of the air-dry soil and sand, measured without any tapping or packing, were placed in a rectangular galvanized sheet-iron pan, 110 x 65 x 20 cm., and were thoroughly mixed. A sufficient quantity of this mixture was prepared at one time to provide for all the cultures to be conducted during the same time period. This procedure secured an approximately uniform soil medium for all the cultures and yielded a soil possessing a water-holding capacity of 28.2 per cent (average of several determinations) of its dry weight, determined according to the method of Hilgard (5).

The containers employed for the soil cultures consisted of stone-ware jars, glazed inside and outside, each with a capacity of 4 liters, and with an inside diameter of approximately 16 cm.

Each soil culture was prepared separately. Four kilograms of the air-dry soil-sand mixture was placed in a mixing pan of galvanized sheet iron. To the dry soil was now added 500 cc. of solution which was then thoroughly mixed with the soil, after which the preparation was placed in a container and the whole weighed to the nearest gram..

The addition of 500 cc. of solution to each 4 kgm. of air-dry soil produced a soil moisture content approximately 12.5 per cent of the weight of air-dry soil, or 44.3 per cent of the water-holding capacity of the soil.

The soybeans used were of the "Wilson" variety. The seed was obtained from the New Jersey Agricultural Experiment Station, having been raised on the experimental grounds of the botanical department of the station. Seeds selected for uniformity of size were germinated in moist sand contained in galvanized sheet-iron pans. The pans stood in the experiment greenhouse until the seedlings were about 4 cm. tall. Vigorous seedlings, nearly equal in size, were selected, carefully removed from the moist sand, and transplanted to the pots containing the soil cultures previously prepared. Five seedlings were thus transplanted to each pot.

To prevent loss of water by evaporation during the growth period, each pot was sealed by pouring over the surface of the soil around the seedlings, melted paraffine wax prepared according to the formula of Briggs (2) and Shantz. The melting point of the wax used was about 40°C. For the purpose of watering

the plants, a paper funnel, partially buried in the soil in the inverted position, was placed in each pot. The funnel extended about one-fourth the distance to the bottom of the pot, its upper end projecting through the wax seal at the soil surface. These funnels, approximately 8 cm. long, with openings of 5 cm. and 1.5 cm. in diameter, were made of heavy wrapping paper and were then thoroughly saturated with melted paraffine to render them impervious to moisture. The upper opening of each funnel was kept tightly closed with a paraffined cork stopper, except for a short period at the time of each watering. Each culture was weighed immediately preceding and directly following the placing of the funnel and the application of the wax seal. The difference in weight was added to the original weight of the container.

The plants were watered every second day during the early stages of growth, and every day during the later stages. At each watering the cultures were weighed and a sufficient amount of distilled water was poured through the funnel of each culture to restore it to its original weight.

At the end of the growing period the plants were cut just above the wax seal. The tops from each culture were placed in a weighing bottle and dried to constant weight at a temperature of about 103°C., and the dry weight obtained.

4. The water cultures

The solutions which were employed in the preparation of the soil cultures above described, were used also in experiments with water cultures. The solutions were prepared according to the formulae given in tables 2 and 3, and were used without alteration, except that to each solution was added a trace of iron in the form of ferric phosphate.

The containers employed consisted of pint "Mason" jars which had previously been thoroughly cleaned with bichromate-sulfuric acid cleaning solution and rinsed with distilled water. With each culture 500 cc. of solution were employed. The soybeans used were of the same variety and were obtained from the same lot as were those used in connection with the soil cultures. The seeds were germinated in moist sphagnum. Selected seedlings about 5 cm. tall were mounted in thoroughly paraffined flat cork stoppers in a manner similar to that described by Tottingham (14, p. 173–175). The stoppers were then placed in the culture jars. Each culture comprised three plants. To exclude the light from the roots of the plants, the culture jars were covered with cylindrical paper shells which were black on the inside and nearly white on the outside. These shells were prepared in a manner similar to that described by the writer (12, p. 344). The solutions were renewed every four or five days.

At the end of the growth period the tops were severed from the roots just at the lower surface of the cork stoppers. The tops were then dried and the dry weights obtained in the same way as were the yields from the soil cultures.

During the periods of growth the general character of the development of the tops in the soil cultures, as well as of the tops and roots in the water cul-

tures, was noted. The characteristics of the injury sustained by the plants of the various culture groups were studied, and the injured plants were compared from time to time with others of the same group and also with the injured plants of the other groups.

EXPERIMENTAL RESULTS

I. Introductory

In the following sections are presented the results of three distinct experiments, each experiment consisting of two series of cultures, of which the second series is a repetition of the first. For the sake of convenience, the two series of each experiment will be designated series A and series B throughout. A comparison will be given of the physiological effects upon the growth of soybeans, produced by the various cultures (of each series) containing the soluble phosphate salts: (a) when these salts were added singly to soil cultures; (b) when added to soil cultures in nutrient mixtures, and (c) when employed in nutrient solutions as water cultures. The responses of the plants to the media in which they grew will be studied with reference to the total yields of dry tops, and especially with respect to the toxic influences of the various salts resulting in any unusual pathological conditions, as these might be detected by general observation.

II. Experiment I. Monobasic phosphate salts singly in soil cultures

In this experiment five groups of cultures were employed, the five groups corresponding to the five different salts, NaH_2PO_4, $(NH_4)H_2PO_4$, KH_2PO_4, $CaH_4(PO_4)_2$, and $MgH_4(PO_4)_2$. Each group comprised 11 cultures, as previously described, including one check culture which received no salt. The substratum in which the plants grew was prepared by thoroughly mixing 500 cc. of pure solution of the salt in question with 4 kg. of air-dry soil-sand mixture. The total amount of salt received by each of these cultures was included in this initial application of 500 cc. of solution. The approximate osmotic values (atm.) of the ten solutions of each series added to the soil cultures, their volume-molecular concentrations, and the weight of the salt in each culture expressed in per cent of the weight of the air-dry soil are given in table 1. Series A of this experiment was conducted from August 25 to October 4, 1916; series B was carried out between February 24 and April 5, 1917.

1. Dry weights. The numerical data of the yields of soybean tops from the cultures of these two series are presented in table 4. The table gives the dry weights of tops for each of the five groups of cultures of series A and of series B. The dry-weight yield for each culture represents the average for six plants. The first column of the table gives the approximate osmotic concentration values of the solutions applied to the cultures. Then follow five sec-

tions with three columns in each section, referring to the dry weights of tops, each section giving the data for a single group. The first two columns in each section present the data for series A and B, respectively, and the third column in each section gives the value obtained by averaging the corresponding data from series A and B. Each of these averages thus represents the data of the two series combined. Each of the dry-weight values is expressed in terms of the corresponding value of the check culture considered as unity, but the actual dry weight of this culture is given in parenthesis in grams, so that the actual weight for any culture may be obtained by multiplying its relative weight by the actual weight of the check culture. The actual average values of the check cultures for series A and B, as given in the first two columns of

TABLE 4

Relative dry weights of soybean tops grown 40 days in soil cultures prepared with pure solutions of monobasic phosphate salts of various concentrations: Series A, conducted from August 25 to October 4, 1916; Series B, from February 24 to April 5, 1917

OSMOTIC CONCENTRATION	GROUP I (NaH_2PO_4)			GROUP II ($NH_4H_2PO_4$)			GROUP III (KH_2PO_4)			GROUP IV [$CaH_4(PO_4)_2$]			GROUP V [$MgH_4(PO_4)_2$]		
	Series A	Series B	Average	Series A	Series B	Average	Series A	Series B	Average	Series A	Series B	Average	Series A	Series B	Average
atm.															
0.0	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
	5.91	7.57	6.74	5.91	7.57	6.74	5.91	7.57	6.74	5.91	7.57	6.74	5.91	7.57	6.74
0.5	0.71	1.01	0.86	0.87	0.73	0.80	0.95	0.93	0.94	0.82	0.80	0.81	0.73	0.96	0.85
1.0	0.59	0.95	0.77	0.85	0.68	0.76	0.85	0.87	0.86	0.81	0.78	0.80	0.70	0.95	0.83
1.5	0.46	0.91	0.69	0.73	0.56	0.66	0.78	0.80	0.79	0.35	0.75	0.55	0.65	0.73	0.69
2.0	0.31	0.79	0.55	0.50	0.41	0.46	0.76	0.77	0.77	0.31	0.58	0.45	0.64	0.53	0.59
2.5	0.26	0.62	0.44	0.32	0.41	0.37	0.75	0.58	0.67	0.30	0.54	0.42	0.48	0.45	0.47
3.0	0.25	0.48	0.37	0.27	0.37	0.32	0.57	0.49	0.53	0.27	0.53	0.40	0.48	0.37	0.43
3.5	0.25	0.42	0.34	0.22	0.28	0.25	0.57	0.43	0.50	0.25	0.41	0.33	0.37	0.35	0.36
4.0	0.23	0.33	0.28	0.18	0.28	0.23	0.54	0.36	0.45	0.22	0.32	0.27	0.32	0.34	0.33
5.0	0.19	0.26	0.23	0.16	0.18	0.17	0.43	0.36	0.40	0.11	0.28	0.20	0.28	0.34	0.31
7.0	0.13	0.24	0.19	0.15	0.15	0.15	0.36	0.35	0.36	0.05	0.26	0.16	0.14	0.23	0.19

each section in the table, were obtained by averaging the yield values of the check cultures from the five groups.

The average relative dry-weight values of each group form a somewhat uniformly decreasing series of numbers. These were plotted to form the graphs of figure 1. Here the ordinates represent the relative dry-weight values, while the abscissas represent the approximate osmotic concentration values (atmospheres) of the single-salt solutions added to the cultures of each group. Since the osmotic concentrations of the solutions added to corresponding cultures of the five groups were approximately the same, the average dry weight values for the five groups were plotted with the same abscissas. The average relative yield from the check cultures is here represented graph-

ically by a horizontal broken line and the actual value in grams is given at the right.

The relative degrees of injury sustained by the plants of the various cultures of each group are graphically represented in figure 1 by the broad vertical lines just below the graph representing the yields of the group in question. The shortest vertical line represents slight injury, vertical lines of medium length indicate pronounced injury, while the longest lines denote severe injury. The relative terms, *slight, pronounced*, and *severe*, will be defined later in connection with the description of the injury produced by the salts.

From the graphs of figure 1 and from the data of table 4, it is at once clear that the growth of soybean tops was very unfavorably influenced by each of the monobasic phosphate salts applied singly to the soil in which the plants were rooted. Each of the graphs slopes uniformly downward to the right, denoting a decrease in the yield of dry tops with each increase in the application of the salt.

As indicated by these graphs, the average yield from the check cultures was considerably higher than that of the highest average yield from any culture of the five groups here employed. Relative to the average yield from the check cultures considered as 1.00, the highest average yield from each of the five series is as follows: series I [NaH_2PO_4], 0.86; series II [$(NH_4)H_2PO_4$], 0.80; series III [KH_2PO_4], 0.94; series IV [$CaH_4(PO_4)_2$], 0.81; series V [$MgH_4(PO_4)_2$], 0.85. The lowest average yields from the five series, taken in the same order as given above are: 0.19, 0.15, 0.36, 0.16, and 0.19. Group III here shows the highest average relative dry-weight values throughout the entire series. The average yields from corresponding cultures of group I and group V are nearly equal and are considerably lower in value than are the average dry weights from corresponding cultures of group III, showing the highest average yields, while group II shows the lowest average yields. It is thus clear that mono-potassium phosphate, aside from any specific injury to the plants, has the least unfavorable influence upon the growth of soybean tops, while mono-ammonium phosphate is the most unfavorable of the five salts, as these were here employed.

2. Injury. As was to be expected, the growth of the plants in those cultures of each group to which were applied solutions having concentrations with osmotic values between 2.5 and 7.0 atmospheres, was soon visibly retarded. This retardation in growth was followed by specific injury to the plants, appearing first in the cotyledons in the form of dark brown discolorations around the margins. In severe injury the discoloration spread rapidly over the whole surface of the cotyledons, and this was followed soon after by death and falling of the organs. The injury next appeared on the foliage leaves. Usually an interval of several days elapsed between the time when the injury first became apparent on the cotyledons and the time of its appearance on the foliage leaves. Injury to the leaves appeared first on or near the margins, in the form of small, yellowish, round spots, which rapidly assumed the

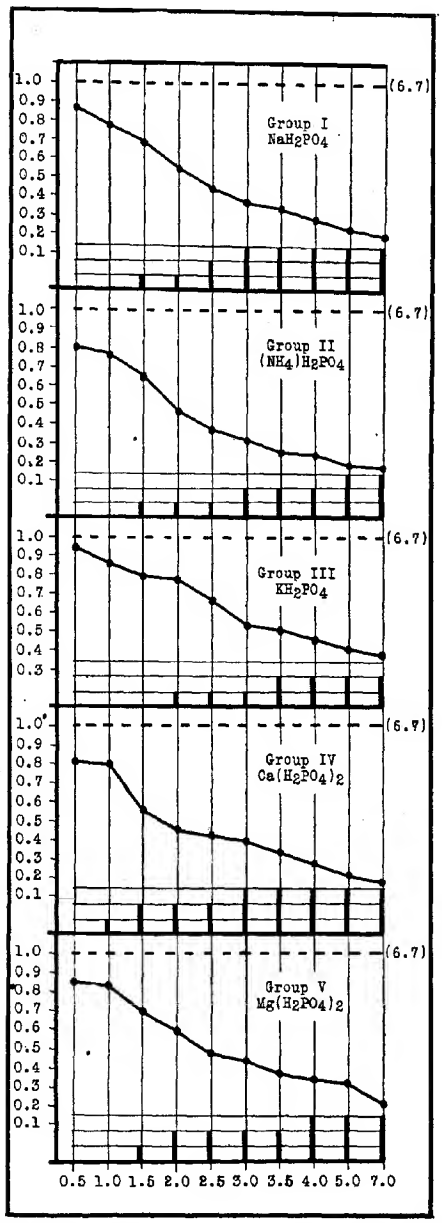

FIG. 1. RELATIVE DRY WEIGHTS AND INJURY OF SOYBEAN TOPS GROWN IN SOIL CULTURES CONTAINING PHOSPHATES SINGLY

characteristic dark brown coloration. In cases of severe injury the affected portions of the leaves gradually increased in size until the injury had spread to the entire leaf surface. This kind of injury was invariably followed by death and falling of the leaves affected. The injury spread, in severe cases, to include all the leaves of the plant. In the later stages of growth, the stems also were affected. This occurred frequently when the leaves of the affected plant were only slightly injured. Injury to the stem appeared in the form of oblong brown spots which gradually increased in size, with shrinking of the tissues.

The cultures of each group may be divided according to the degrees of injury sustained, into four classes: (a) those without specific injury; (b) those which exhibited *slight* injury, not sufficiently severe to cause falling of the leaves affected; (c) those which sustained *pronounced* injury, severe enough to cause the leaves to fall but not sufficiently severe to cause the death of the plants before the end of the growth period; (d) those which sustained *severe* injury resulting in the death of the plants before the end of the growth period. As previously stated, the relative degrees of injury are graphically represented in figure 1 by broad vertical lines just below the graph representing the dry-weight yields of the series in question.

A comparison of the diagrams representing the relative degrees of injury sustained by the cultures of the various groups, brings out the fact that the total injury exhibited by group III [KH_2PO_4] was less than that sustained by any of the other groups. This group showed four cultures with pronounced injury and three with slight, leaving three uninjured cultures. Group IV [$CaH_4(PO_4)_2$], on the other hand, exhibited the greatest total injury of any group. As indicated by the diagram, five cultures in this group were killed before the end of the growth period, three sustained pronounced injury, and one exhibited slight injury, leaving only one uninjured culture in the entire group. Each of the remaining three groups (I, II and V) possessed two cultures which remained uninjured throughout the entire period. Groups I and IV each possessed five cultures which sustained severe injury resulting in the death of the cultures, and these were corresponding cultures of the two groups.

From these considerations it is clear that the salt KH_2PO_4, (group III), as here used, is less pronounced in its capacity to produce specific injury to soybean tops than are any of the other salts here employed. Mono-calcium phosphate (group IV), on the other hand, shows a more pronounced harmful influence with respect to the injury observed, than do any of the other salts.

A careful comparison of the injured plants showed that the nature of the injury sustained by the cultures of the various groups was identical. The only apparent differences, apart from size, between the plants from the cultures of the different groups were the degrees of the injury sustained. It is to be concluded, therefore, that the injury to the plants resulting from the presence of the phosphate salts, as these were here employed singly, was due to

some property common to all the solutions used. The injury cannot be the result of mere physical concentration, since it occurred in plants of those cultures to which were applied solutions having concentrations with no greater osmotic values than from 1 to 2.5 atmospheres: concentrations which have repeatedly been shown to be not above the optimum for good plant growth. Moreover, the concentrations of these solutions cannot be supposed to remain unchanged upon being introduced into the soil, since the phosphates when applied to the soil in soluble forms are undoubtedly, to a large extent at least, eventually precipitated in combination with various bases existing in the soil. That the injury is accentuated by excessive concentration, is clearly evident from the graphs of figure 1.

It thus appears that the specific injury here observed can not be attributed to the influence of the cations, for it is scarcely to be supposed that each of the five ions, Na, NH_4, K, Ca and Mg, could affect the plants alike in this respect. The injury, therefore, must be related to the common atomic group H_2PO_4, or to the ions resulting from the partial dissociation of this group in the soil solution. This conclusion is in harmony with that of Harris (4) who found that to the anion more than to the cation, is to be attributed the toxic effects of the alkali salts in the soil. That the H-ion concentration of these monobasic phosphates singly in solutions is comparatively high, is of course, well understood, and the injury here observed is undoubtedly related to the H-ion concentration in the soil solution.

III. Experiment II. Monobasic phosphate salts in soil cultures with complete nutrient mixtures

In this experiment six groups of cultures were employed, corresponding to the six series of mixed solutions whose formulas are presented in tables 2 and 3. The tables give the approximate partial volume-molecular concentrations of the solutions as these were here used. Each solution of the six series had a total osmotic concentration value of approximately 2.5 atmospheres. As described in a former section, the soil cultures of each group were prepared by mixing separately 500 cc. of each solution of the series in question with 4 kgm. of air-dry soil. The total amount of salts added to each culture was included in this initial application of 500 cc. of the mixed solution. Each group comprised 21 cultures, including one check culture, which was prepared in the same way as were the other cultures, except that the dry soil received an initial application of 500 cc. of distilled water instead of solution.

During the growth period the cultures stood in parallel rows (20 cultures in a row) on central tables in the experimental greenhouse, and were frequently shifted in their positions according to a definite plan. Series A of this experiment was conducted from November 5 to December 5, 1916. Series B was exactly like series A, but was carried out between January 10 and February 9, 1917.

1. Dry weights. The numerical data of the yields of tops are presented in table 5. The table comprises six sections, each of which refers to the data of a single group. The first two columns in each of the six sections give the dry weights of tops relative to the average dry weight of the six check cultures for series A and B, respectively. The third column of each section gives the average dry-weight yield for series A and series B together. The average absolute dry weights of the check cultures for series A and B are given in parenthesis. The culture numbers given in the first column of the table refer to the positions occupied in the Tottingham series by the solutions employed in the preparation of the soil cultures.

To bring out the relation between the salt proportions and the relative dry yields of tops of the various groups, the data of table 5 are graphically represented in figure 2. To prepare these graphs, the average relative dry weights of tops of the 3-salt group were first arranged in the order of their magnitudes, beginning with the highest, to form a somewhat uniformly decreasing series of numbers. The relative yields of each of the other five groups were then arranged in the same order, after which they were plotted to form the five graphs of figure 2, shown as the irregular dotted lines. The abscissas were here arbitrarily chosen to represent the cultures, the numbers of which, being the same for corresponding cultures of each of the five groups, are placed below. The ordinates represent the average relative dry-weight values. The average relative yield from the check culture is represented by a horizontal broken line, and the actual average value in grams is given at the right. As in figure 1, the broad vertical lines just below each set of graphs indicate the relative degrees of injury sustained by the plants of the cultures indicated. The numbers at the top indicate the number of tenths of the total osmotic concentration due to the total phosphates in the solutions whose numbers are given below.

In order to bring out more clearly the effect upon the growth of soybean tops of an additional application of the monobasic phosphate salts to soil cultures containing a complete fertilizer, the graph representing the average relative dry weights of tops of the 3-salt group, arranged in the descending order of their values, was superimposed upon each of the other graphs, using the same scale of ordinates. This graph exhibits a uniformly downward slope to the right, as was previously arranged.

Each graph of figure 2 has a decided tendency to slope downward to the right, but the graph of each group except group V crosses the graph of the 3-salt group at various points, but chiefly in its lower portions, thus indicating that many of the poorer 3-salt proportions were improved for the growth of soybean tops by the presence of the additional monobasic phosphate salt. The best set of salt proportions of the 3-salt group, however, was decreased in productiveness by the presence of each one of the phosphate salts except monocalcium phosphate (group IV). The maximum average yield of group IV slightly exceeds that of the 3-salt group, the former having a value of 1.63 as compared with the average yield of the check cultures, while the latter has a

Relative dry weights of soybean tops grown thirty days in soil cultures prepared with mixed solutions having total osmotic concentration values of approximately 2.5 atmospheres. Series A, conducted from November 5 to December 5, 1916; Series B, from January 10 to February 9, 1917

CULTURE NUMBER	GROUP I			GROUP II			GROUP III			GROUP IV			GROUP V			3-SALT GROUP		
	Series A	Series B	Average	Series A	Series B	Average	Series A	Series B	Average	Series A	Series B	Average	Series A	Series B	Average	Series A	Series B	Average
Check	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
	2.12	2.76	2.442	2.12	2.76	2.442	2.12	2.76	2.442	2.12	2.76	2.442	2.12	2.76	2.442	2.12	2.76	2.442
$T_1R_1C_1$	0.97	1.36	1.17	1.08	1.21	1.15	1.32	1.10	1.21	1.61	1.18	1.40	1.33	1.10	1.22	1.69	1.09	1.39
C_3	1.02	1.38	1.20	1.56	1.07	1.32	1.20	1.01	1.11	1.67	1.21	1.44	1.17	1.04	1.11	1.54	1.08	1.31
C_5	1.05	1.28	1.17	1.52	1.20	1.36	1.32	0.93	1.13	1.81	1.07	1.44	1.25	0.85	1.05	1.35	0.96	1.16
C_7	1.19	1.36	1.28	1.54	1.97	1.26	1.40	0.94	1.17	1.63	0.79	1.21	0.60	0.84	0.72	1.08	0.91	1.00
R_3C_1	1.68	1.54	1.60	1.67	1.35	1.51	1.68	1.30	1.49	1.99	1.27	1.63	1.68	1.10	1.39	1.84	1.37	1.61
C_3	0.98	1.37	1.18	1.49	1.30	1.40	1.39	1.03	1.21	1.72	1.15	1.44	1.25	1.03	1.14	1.56	1.10	1.33
C_5	1.32	0.93	1.13	1.34	1.17	1.26	1.08	0.95	1.02	1.72	1.06	1.39	0.80	0.92	0.86	1.53	1.00	1.27
R_5C_1	1.24	1.36	1.30	1.65	1.20	1.43	1.13	1.01	1.07	1.43	1.31	1.37	1.14	1.01	1.08	1.56	1.11	1.34
C_3	1.12	1.42	1.27	1.53	1.18	1.36	1.41	0.93	1.17	1.50	1.18	1.34	1.16	0.99	1.06	1.58	1.08	1.33
R_7C_1	1.24	1.36	1.30	1.47	1.34	1.41	1.24	1.01	1.13	1.03	1.21	1.12	1.43	1.04	1.24	1.51	1.16	1.34
$T_3R_1C_1$	1.09	1.24	1.17	1.75	1.18	1.47	1.43	1.17	1.30	1.67	1.17	1.42	1.37	1.04	1.21	1.41	1.13	1.27
C_3	1.49	1.33	1.41	1.13	1.07	1.10	0.94	1.13	1.04	1.44	1.04	1.24	0.98	0.93	0.96	1.31	1.08	1.20
C_5	1.12	1.32	1.22	1.56	1.04	1.30	0.93	0.92	0.93	1.47	0.87	1.17	1.01	0.93	0.96	0.94	0.99	0.97
R_3C_1	1.26	1.16	1.21	1.43	1.23	1.33	1.13	1.18	1.16	1.34	1.17	1.26	1.17	1.01	1.09	1.79	1.21	1.45
C_3	1.16	1.38	1.27	1.62	1.01	1.32	1.48	1.14	1.31	1.29	1.09	1.19	1.25	1.03	1.14	1.32	1.06	1.19
R_4C_1	0.91	1.27	1.09	1.43	1.11	1.27	1.31	1.05	1.18	1.10	0.85	0.98	1.00	1.00	1.00	1.33	1.04	1.19
$T_5R_1C_1$	1.10	1.26	1.18	1.56	1.12	1.34	1.36	1.11	1.24	0.87	0.88	0.88	1.15	1.05	1.10	1.69	1.19	1.44
C_3	1.16	1.20	1.18	1.52	0.91	1.22	1.07	1.00	1.04	1.16	0.88	1.02	1.06	0.93	1.00	1.23	0.92	1.08
R_3C_1	1.00	1.15	1.08	1.59	1.12	1.36	1.45	1.18	1.32	0.93	0.95	0.94	0.88	0.88	0.88	1.29	0.85	1.07
$T_7R_1C_1$	1.04	1.04	1.04	1.05	0.97	1.01	1.26	1.04	1.15	0.94	0.44	0.69	0.80	0.83	0.82	1.20	0.80	1.00

corresponding value of 1.61. It will be observed that the graph representing yields for group V lies below the graph of the 3-salt group throughout its en-

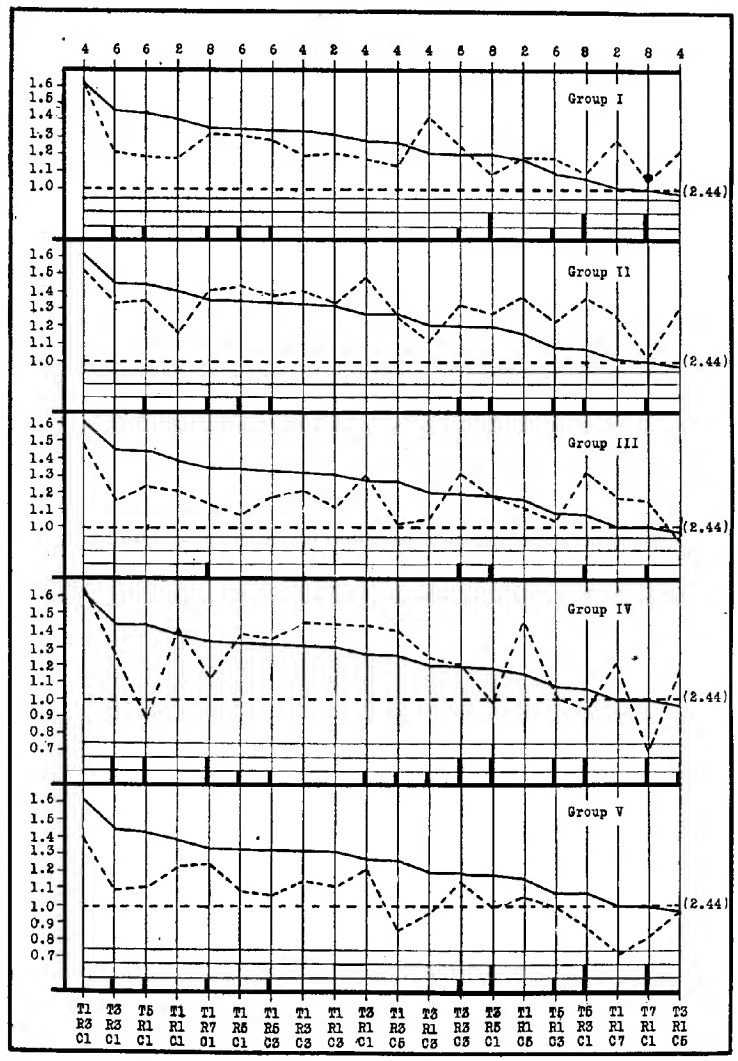

FIG. 2. RELATIVE DRY WEIGHTS AND INJURY OF SOYBEAN TOPS GROWN IN SOIL CULTURES CONTAINING PHOSPHATES IN CONNECTION WITH A COMPLETE FERTILIZER RATION

tire length. It is thus clear that each set of salt proportions of the 3-salt group is decreased in productiveness by the presence of mono-magnesium phosphate.

The graph representing the average yield from the 3-salt group shows clearly enough that the soil here employed was improved for the growth of soybeans by the application of all but three of the various sets of salt proportions of the three salts, KH_2PO_4, $Ca(NO_3)_2$ and $MgSO_4$, in solutions having total osmotic concentration values of 2.5 atmospheres, when the applications of the solutions were made to yield a soil moisture content of 12.5 per cent on the basis of the weight of the air-dry soil (500 cc. of solution to each 4 kgm. of soil). The maximum average yield of the 3-salt group was obtained from culture $T_1R_3C_1$. The yield from this culture showed an improvement of 61 per cent over that of the average yield from the check cultures, while the minimum average yield, obtained from culture $T_3R_1C_5$, was 3 per cent below the average yield from the checks. The salt proportions of the solutions added to these two cultures are: KH_2PO_4, 0.0173 m.; $Ca(NO_3)_2$, 0.0040 m.; $MgSO_4$, 0.0322 m., for the former, and KH_2PO_4, 0.0074 m.; $Ca(NO_3)_2$, 0.0257 m.; $MgSO_4$, 0.0083 m., for the latter. With other total concentrations of the solutions than the one here employed, it may be expected that the salt proportions required to produce maximum growth would be different from the ones here noted, since it has been pointed out by several authors (3, 14, 12, 8, 9, 1) that physiological balance of salt proportions in nutrient media is markedly dependent upon total concentration. It is thus clear that many degrees of beneficial effects and some injurious ones upon the growth of soybeans may be obtained by the application of various proportions of the three salts, when the total quantities of the three salts in solution applied to each soil culture, have approximately equal osmotic values. This emphasizes the importance of considering, not only salt proportions, but also total quantities applied (concentration), in the use of fertilizer salts in agricultural practice.

The graphs representing the yields of group I (NaH_2PO_4) and group II [$(NH_4)H_2PO_4$] show that all the average yields from the cultures of these groups were above the average yield from the check cultures, while groups III, IV, and V show one, four, and five cultures, respectively, which produced lower average yields than did the checks. As is thus clearly indicated, the soil employed in these cultures was improved for the growth of soybean tops by each set of salt proportions of groups I and II, and by all but one set of salt proportions of group III, while four sets of salt proportions of group IV, and five sets of group V, reduced the productiveness of this medium.

Further inspection of the graphs of figure 2 and also of the data of table 5 brings out the fact that in each of the five groups the highest average yield occurred with culture $T_1R_3C_1$, which corresponds also with the culture of the 3-salt group producing the highest yield. The maximum average yields for these five groups, relative to the average yields from the check cultures, are as follows: group I, 1.60; group II, 1.51; group III, 1.49; group IV, 1.63; group V, 1.39. The maximum yield from the 3-salt group has a corresponding value of 1.61, which is slightly surpassed only by the maximum average yield from group IV, having a value of 1.63.

2. Injury. The cultures of the 3-salt group suffered no specific injury, but remained in an apparently healthy and vigorous condition throughout the entire growth period. The cultures of the other five groups gave no evidence of disturbed growth during the first two weeks after the young plants had been placed in the soil cultures containing the monobasic phosphates in addition to the complete nutrient mixture. The plants showed differences in size, but no unusual phenomena occurred during this time. During the third week however, evidences of disturbed growth began to appear in the plants of several cultures of each group. The cotyledons were the parts first to be injured. The injury appeared in the form of dark brown discolorations around the margins of these organs. Later it appeared on the foliage leaves and in a few cases, on the stems of the plants also. In no culture, however, was the injury sufficiently severe to cause the death of the plants before the end of the growth period.

Careful comparisons revealed no differences between the characteristics of the injury sustained by the plants of the various groups. The injured cultures of these five groups were also compared with those of the preceding experiment, where the media in which the plants were rooted contained the monobasic phosphates singly. Aside from the varying degrees of injury, this comparison showed no apparent differences in the nature of the injury sustained by the cultures of the two experiments. It appears, therefore, that the nature of the toxin producing the specific injury here observed is the same, not only in the cultures of the groups employing the different monobasic phosphates singly, but also in those containing these phosphates in combination with the complete nutrient mixtures.

The injured cultures and the degrees of injury sustained are indicated in figure 2 by short, broad, vertical lines just below the graphs representing the relative yields, in a manner similar to that employed in figure 1. Vertical lines of two lengths only are employed in figure 2, since, as above stated, no severe injury was observed in the cultures of this experiment. The shorter vertical lines indicate slight injury, and the longer ones denote pronounced injury. As previously stated, the numbers at the top indicate the number of tenths of the total osmotic concentration (2.5 atmospheres), due to the total phosphates in the solutions whose numbers are given below.

The diagrams of figure 2 show that the number of injured cultures varied with the different groups as in the preceding experiment. The least total injury occurring in a single group is indicated for group III, with five cultures only slightly injured. The largest number of injured cultures exhibited by a single group occurred in group IV. This group comprised eight cultures which showed pronounced injury and six which were slightly injured, leaving only six uninjured cultures in the entire group. Groups I and V each comprised three cultures which showed pronounced injury and six with slight injury, making a total of nine injured cultures in each group, while group II exhibited the same number of injured cultures, none of which, however, showed pronounced injury.

It will be observed that in every group except group IV, the injury was confined to those cultures, each of which had received an application of solution whose partial osmotic concentration due to the total phosphates was not less than six-tenths of the total concentration of the solution, while pronounced injury occurred only in those cultures prepared with solutions having eight-tenths of their total concentrations due to the phosphates. In group IV, slight injury was observed in four cultures ($T_3R_1C_1$, $T_1R_3C_5$, $T_3R_1C_3$, and $T_3R_1C_5$) having four-tenths, and two cultures ($T_1R_5C_1$, and $T_1R_5C_3$) having six-tenths of their total osmotic concentrations due to the phosphates, while all the remaining cultures prepared with solutions having six-tenths or eight-tenths of their total concentration due to the phosphates, showed pronounced injury. It is thus clear that the $CaH_4(PO_4)_2$ used in the preparation of the cultures of this group has a higher capacity for producing this injury when used in connection with a complete nutrient mixture, than have any of the other phosphate salts here employed.

It is important to note that the cultures of groups II, III, and V, prepared with solutions having six-tenths of their total osmotic concentrations due to total phosphates, did not all suffer injury, nor did all the cultures prepared with solutions whose partial concentrations due to the phosphates were eight-tenths of the total osmotic concentration, sustain pronounced injury. Conditions similar to these prevailed also in group I, and in a more striking manner in group IV, where out of a total number of six cultures prepared with solutions having six-tenths of their total osmotic concentration due to the phosphates, four showed pronounced injury and two showed slight injury, and out of a total number of six cultures prepared with solutions, four-tenths of whose total concentrations were made up of the phosphates, four were slightly injured and two showed no injury at all. It is thus obvious that the degree of the injury sustained by the cultures of this experiment was not determined solely by the total phosphate content of the medium in which the plants were grown, but was largely influenced by the relative proportions of the four salts.

3. Relation of injury to yields. There appears to be no clearly definite relation between the injury here observed and the dry weight of tops, but in a general way pronounced injury is coincident with relatively low yields. Slight injury appears to have no greater tendency to occur with low yields than with high yields. For example, two cultures, $T_3R_3C_3$ and $T_5R_3C_1$, of group III each showed slight injury and gave relatively high yields of tops, while cultures $T_1R_3C_5$ and $T_3R_1C_3$ of the same group remained uninjured but produced relatively low yields. In group II, the cultures $T_1R_1C_1$ and $T_3R_1C_3$, both uninjured cultures, produced low yields, while the slightly injured cultures $T_1R_7C_1$ and $T_1R_5C_1$ each yielded relatively high dry weights of tops. Examples similar to these might be pointed out in each of the five groups. It is clear, therefore, that in the soil cultures here employed, slight injury does not materially interfere with the production of high yields of tops.

IV. Experiment III. Monobasic phosphate salts in solution cultures with complete nutrient mixtures

The solutions used in this experiment were the same as those used in the preparation of the soil cultures of the preceding experiment. The preparation of the solution cultures, the germination of the seeds, and the general treatment of the plants during the growth period have been described in a previous section. The formulas for the solutions of the five four-salt series are presented in table 2, and in a similar manner the composition of the solutions of the three-salt series is given in table 3. Each group thus comprised 20 cultures. An additional culture in Knop's solution with the same total osmotic concentration (2.5 atm.) was added to each group.

The experiment was carried out in the greenhouse with the cultures arranged in rows (one group of cultures in each row) on tables centrally located. In order to expose the cultures as nearly alike as possible to the constantly changing environment, they were regularly shifted in their positions according to a definite plan. Each of the two series of cultures here considered extended over a time period of 24 days. Series A was conducted from January 8 to February 1. Series B, which was a repetition of series A, was carried out between February 7 and March 4, 1917.

1. Dry weights of tops. The dry weight yields of tops expressed in terms of the average yield from Knop's solution taken as 1.00, are presented in table 6. The data in this table are arranged in the same manner as are those in table 5. The actual average yield of tops from Knop's solution is given in parenthesis below the relative value (unity) in each case, so that the actual value of any culture may be calculated. It will be observed that the data for five groups only appear in the table. The data for group IV are here omitted, owing to the fact that the solutions containing $CaH_4(PO_4)_2$ proved entirely unsuited to the growth of soybeans. The plants of most of the cultures were so severely injured that growth was impossible, and the plants of all the cultures except those of cultures $T_1R_1C_1$, $T_1R_1C_3$, $T_1R_1C_5$, and $T_1R_1C_7$, were killed before the end of the growth period. The cultures of this group were not repeated.

As in the case of the yields from the soil cultures of the preceding experiment, the data of table 6 have been graphically represented and these graphs are presented in figure 3. In both cases the data for the 3-salt group of cultures were arranged in the decreasing order of their values and then plotted as ordinates, with the abscissas arbitrarily taken to represent the different cultures. The order of the cultures is thus not the same in figures 2 and 3. After the descending order of values for the 3-salt group was determined, the data for the 4-salt groups were plotted in this order, with the same scale of ordinates in each case, to form the four graphs represented by the irregular dotted lines. The graph representing the yields from the 3-salt group is superimposed upon each of the other graphs for ready comparison. The horizontal dotted line

TABLE 6

Relative dry weights of soybean tops grown 24 days in culture solutions, all with total concentrations having osmotic values of approximately 2.5 atmospheres. Series A, conducted from January 8 to February 1; Series B, from February 7 to March 3, 1917

CULTURE NUMBER	GROUP I			GROUP II			GROUP III			GROUP V			3-SALT GROUP		
	Series A	Series B	Average	Series A	Series B	Average	Series A	Series B	Average	Series A	Series B	Average	Series A	Series B	Average
Knop's solution	1.00 (0.70)	1.00 (0.66)	1.00 (0.679)	1.00 (0.70)	1.00 (0.66)	1.00 (0.679)	1.00 (0.70)	1.00 (0.66)	1.00 (0.679)	1.00 (0.70)	1.00 (0.66)	1.00 (0.679)	1.00 (0.70)	1.00 (0.66)	1.00 (0.679)
$T_1R_1C_1$	1.38	1.45	1.42	1.38	1.14	1.26	0.64	0.62	0.63	0.64	0.50	0.57	1.55	1.28	1.42
C_3	1.50	1.42	1.46	1.19	1.18	1.19	1.15	1.07	1.11	0.62	0.69	0.66	1.43	1.64	1.54
C_5	1.27	1.35	1.31	1.20	1.29	1.25	1.20	1.14	1.17	0.44	0.45	0.45	1.28	1.18	1.23
C_7	1.40	1.37	1.39	0.87	1.04	0.96	0.70	0.72	0.71	0.60	0.56	0.58	0.74	0.91	0.83
R_3C_1	0.60	0.50	0.55	0.45	0.46	0.46	0.40	0.50	0.45	0.40	0.34	0.37	0.44	0.43	0.44
C_3	0.60	0.55	0.58	0.45	0.50	0.48	0.56	0.45	0.51	0.41	0.40	0.41	0.47	0.56	0.52
C_5	0.53	0.55	0.54	0.47	0.55	0.51	0.52	0.50	0.51	0.44	0.42	0.43	0.43	0.52	0.48
R_5C_1	0.39	0.45	0.42	0.32	0.47	0.40	0.46	0.42	0.44	0.44	0.30	0.37	0.36	0.46	0.41
C_3	0.46	0.46	0.46	0.41	0.48	0.45	0.46	0.38	0.42	0.38	0.43	0.41	0.41	0.45	0.43
R_7C_1	0.36	0.32	0.34	0.36	0.37	0.37	0.40	0.41	0.41	0.30	0.34	0.32	0.37	0.38	0.38
$T_3R_1C_1$	1.37	1.46	1.42	1.41	1.45	1.43	0.50	0.46	0.48	0.51	0.43	0.47	1.05	0.98	0.02
C_3	1.16	1.34	1.25	1.32	1.13	1.23	0.53	0.48	0.51	0.44	0.40	0.42	0.86	0.70	0.78
C_5	1.02	1.28	1.15	0.88	1.01	0.95	0.58	0.50	0.54	0.39	0.36	0.38	0.63	0.71	0.67
R_3C_1	0.93	0.55	0.74	0.42	0.50	0.46	0.50	0.41	0.46	0.40	0.52	0.46	0.44	0.47	0.46
C_3	0.74	0.59	0.67	0.42	0.46	0.44	0.41	0.37	0.39	0.40	0.40	0.40	0.42	0.50	0.46
R_5C_1	0.37	0.44	0.41	0.38	0.44	0.41	0.43	0.47	0.45	0.38	0.32	0.35	0.37	0.43	0.40
$T_5R_1C_1$	0.97	1.51	1.24	0.53	1.00	0.77	0.38	0.40	0.39	0.55	0.43	0.49	0.67	0.59	0.63
C_3	0.76	1.37	1.07	0.87	0.96	0.92	0.44	0.42	0.43	0.53	0.41	0.47	0.46	0.51	0.49
R_3C_1	0.82	0.75	0.79	0.44	0.56	0.50	0.45	0.36	0.41	0.33	0.45	0.39	0.36	0.50	0.43
$T_7R_1C_1$	0.66	1.11	0.89	0.58	0.71	0.65	0.50	0.46	0.48	0.42	0.40	0.41	0.48	0.53	0.51

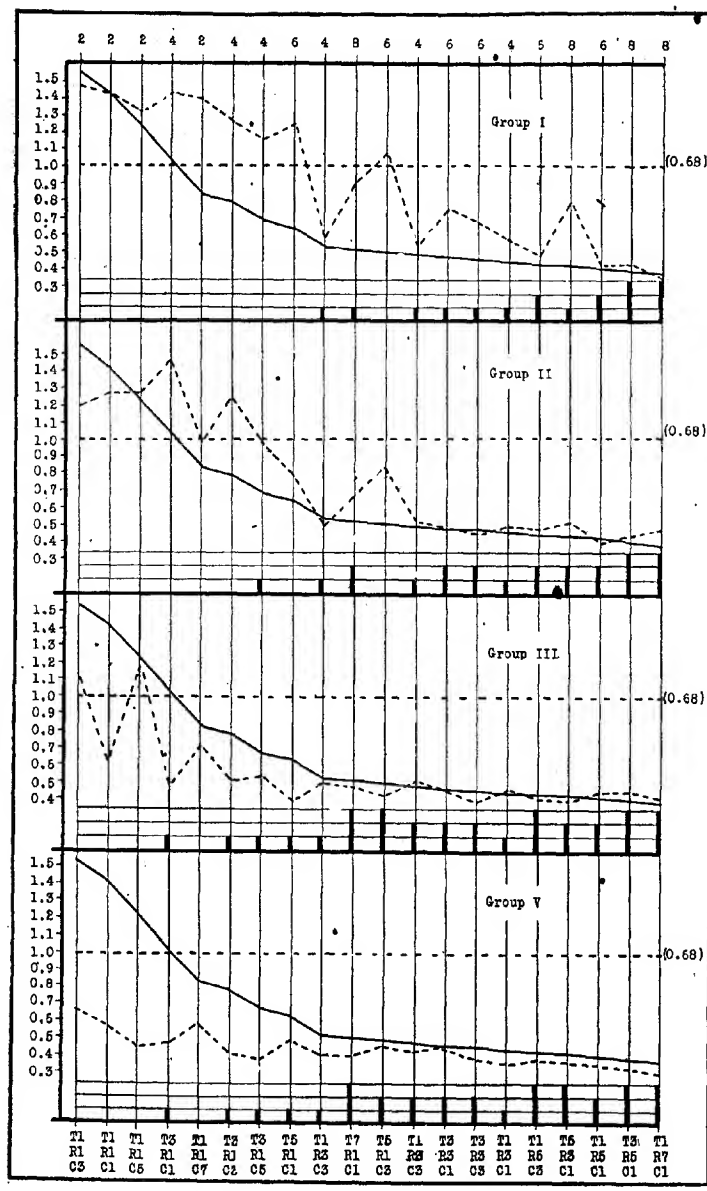

FIG. 3. RELATIVE DRY WEIGHTS AND INJURY OF SOYBEANS GROWN IN SOLUTIONS CONTAINING PHOSPHATES IN CONNECTION WITH COMPLETE NUTRIENT MIXTURES

here represents the average relative value of the yields from Knop's solution. The actual average value of these yields is given in parenthesis at the right. As in figure 2, the degrees of injury sustained by the cultures are represented by the broad perpendicular lines below each graph. The numbers at the top represent, in each case, tenths of the total osmotic concentration due to the total phosphate salts in the solutions whose numbers appear below.

Perhaps the most striking feature brought out by the graphs of figure 3 is the fact that all but two of the yields from the cultures of group I, and all except five of the yields from group II, were markedly higher than were the yields from corresponding cultures of the 3-salt group, while the yields from all but five cultures of group III and all except one culture of Group V were lower than were the yields from corresponding cultures of the 3-salt group. The marked improvement of the yields from many of the cultures of Groups I and II, over the yields from corresponding cultures of the 3-salt group, may perhaps be explained by the fact that with the introduction of NaH_2PO_4 into the solutions of group I, and $NH_4H_2PO_4$ into the solutions of group II, a new ion was introduced into the solutions of each of these two groups: Na ions into the solutions of the former and NH_4 ions into the solutions of the latter. Here, possibly, the antagonistic effects of the new ions served to improve the media for the growth of soybeans in the general manner suggested by Osterhout (10). On the other hand, no new ions were added to the solutions of groups III and V with the introduction of the additional KH_2PO_4 into the solutions of the former and $MgH_4(PO_4)_2$ into those of the latter. The ions introduced into the solutions with these two salts served merely to increase the partial concentrations and to change the proportions of the ions given to the solutions by the three salts KH_2PO_4, $Ca(NO_3)_2$ and $MgSO_4$.

It is important to note, however, that the maximum yield from the 3-salt group is not equaled by the maximum yield from any of the other groups. The highest yield from the 3-salt group occurred with culture $T_1R_1C_3$, having salt proportions as follows: KH_2PO_4, 0.0058 m.; $Ca(NO_3)_2$, 0.012 m.; and $MgSO_4$, 0.0323 m. The average yield from this culture had a dry-weight value of 1.54, relative to the average yield from Knop's solution taken as unity. This culture corresponded also to the culture ($T_1R_1C_3$) in group I and in group V producing maximum yields, while the highest yields in groups II and III occurred with cultures $T_3R_1C_1$ and $T_1R_1C_5$, respectively. The maximum average yields from groups I, II, III, and V were 1.46, 1.43, 1.17 and 0.66, respectively, relative to the average yield from Knop's solution.

2. *Injury.* While only the two cultures $T_1R_7C_1$ and $T_3R_5C_1$ of the 3-salt group were severely injured, the four cultures $T_1R_5C_1$, $T_5R_3C_1$, $T_1R_5C_3$ and $T_3R_3C_1$ showed pronounced injury, and the four cultures $T_1R_3C_1$, $T_3R_3C_3$, $T_7R_1C_1$ and $T_1R_3C_3$ sustained slight injury. The total injury sustained by the cultures of this group, however, was less than that of any of the other groups. The uninjured cultures made rapid and vigorous growth.

The interval of time which elapsed after the seedlings had been placed in

the solutions until the injury became apparent in the cotyledons of the seedlings, was very short. In two cultures ($T_1R_7C_1$ and $T_3R_5C_1$) of each group except the 3-salt group, the plants showed signs of specific injury on the second day after they had been placed in the solutions. All the cultures which remained uninjured at the end of 12 days suffered no injury at all during the remainder of the growth period, and some which had been only slightly injured completely recovered and made excellent growth. The time required for the appearance of the injury in the solution cultures here employed is thus in direct contrast with the time required for its appearance on the plants of the soil cultures of the preceding experiment, where no evidences of injury became apparent during the first two weeks of the growth period.

Careful comparisons were made between the injured cultures of the various groups in order to determine whether the characteristics of the injury differed in any respect with the cultures of the different groups, but no differences in the nature of the injury could be detected. Nor did comparisons of the plants in the water cultures of the present experiment with those of the soil cultures of the preceding experiments reveal any apparent difference in the nature of the injury to the cultures of the experiments here considered. It thus appears that the specific injury to the soybean plants of these experiments is directly related to some property which is common to the solutions employed in the preparation of all the cultures which show this injury.

Turning now to the diagrams of figure 3, representing the relative degrees of injury of the four groups, it will be observed that the number of injured cultures varies with the different groups, the smallest number of injured cultures being indicated for group I. This group comprises seven cultures which were slightly injured, two which showed pronounced injury, and two which suffered severe injury, making a total of eleven injured cultures for the entire group, leaving nine cultures which remained uninjured. Group II showed four cultures which sustained slight injury, six with pronounced injury, and two which suffered severe injury, leaving eight uninjured cultures in the group. Groups III and V each exhibited sixteen injured cultures, leaving only four uninjured cultures in each group. Six cultures of each of these two groups showed slight injury, five sustained pronounced injury, while five suffered severe injury. The number of uninjured cultures in groups III and V is thus seen to be only half the number in group II, and less than half the number in group I.

No injury was produced in any of the four groups by solutions whose partial concentration due to the total phosphate was not greater than two-tenths of the total concentration of the solution, although in groups III and V all the solutions with higher partial osmotic concentration of the phosphates produced the injury. In both group I and group II, however, the two solutions $T_3R_1C_1$, and $T_3R_1C_3$, with four-tenths, and solutions $T_5R_1C_1$, and $T_5R_1C_3$, with six-tenths of their total concentrations due to the phosphates, produced no injury.

In the four groups of cultures here considered, it is obvious that not only do the degrees of injury sustained by the plants vary with the variations in the partial osmotic concentrations of the total phosphates, but also with variations in the proportions of the four salts. This is well brought out in group I, where, out of a total number of six cultures with solutions having partial osmotic concentrations due to total phosphates, equal to six-tenths of the total concentrations of the solutions, two cultures showed pronounced injury, two sustained slight injury, and two remained uninjured. In the same group, out of a total number of four solutions with eight-tenths of their total osmotic concentrations due to the phosphates, two solutions, $T_3R_5C_1$ and $T_1R_7C_1$, produced severe injury, while the remaining two solutions, $T_7R_1C_1$ and $T_5R_3C_1$, produced only slight injury. Of the total number of seven solutions of this group which produced slight injury, two had eight-tenths, two had six-tenths, and three had four-tenths of their total osmotic concentrations due to the total phosphates in the solutions. Similar relations of injury to salt proportions and to partial osmotic concentrations due to the total phosphates in the solutions, are clearly indicated also in each of the groups II, III and V. This emphasizes the point already brought out in connection with a similar study of the corresponding groups of soil cultures of the preceding experiment, that the degree of injury sustained by the various cultures is not determined solely by the total phosphate content of the medium in which the plants are grown, but is related also to the proportions of the salts.

3. Injury to roots. Frequent inspection of the roots of the various cultures during the first few days of the growth period, revealed the fact that in those solutions which produced pronounced and severe injury to the tops, the roots were checked in their development soon after being placed in the solutions, and showed specific injury even before disturbed growth in the tops became apparent. At the end of the first week after the seedlings had been placed in the solutions, the injured root systems were characterized by a general unhealthy appearance and by the suppression of lateral root development. Microscopical examination at the end of a week's growth showed the root tips of the more severely injured cultures to be dead, and some of the cells of the root tips to be in a state of disintegration. The roots of the cultures thus affected gradually turned brown in color and the roots of the two cultures $T_1R_7C_1$ and $T_3R_5C_1$ of each group, with the roots of cultures $T_7R_1C_1$, $T_5R_1C_3$, and $T_1R_5C_3$ from group III, and those of cultures $T_7R_1C_1$, $T_1R_5C_3$, and $T_5R_3C_1$ from group V, were killed before the end of the growth period. This latter condition of the roots was always coincident with severe injury to the tops. In a few of the cultures ($T_7R_1C_1$, $T_3R_3C_1$, and $T_5R_3C_1$ of group I, and $T_3R_1C_5$ of group II), which were characterized by slight leaf injury, the roots appeared to be especially well developed, showing evidences of stimulated root growth, while most of the cultures thus characterized showed root development which was somewhat below the average. In cultures where no leaf injury at all was apparent, root development was excellent, but not quite so vigorous as in those cultures above

indicated which suffered slight injury but also gave evidence of stimulated root growth.

4. Relation of injury to yields. In order to facilitate the study of the relations between the injury here observed and the average relative yields of tops, reference may be had to the graphs and diagrams of figure 3. It is to be observed that the lowest average yields occurred with those cultures which suffered either pronounced or severe injury, while the highest average yields were obtained from the cultures which were free from injury. This relation is clearly definite for each group of cultures. In general it may be said that those cultures of each group which suffered slight injury produced relatively low yields, while medium yields were obtained from cultures without injury.

Variations in the yield values with variations in the degrees of the injury sustained, were comparatively slight, since all the cultures which suffered severe, pronounced or slight injury produced relatively low yields. In these cultures, therefore, the injury appears to be the controlling factor in the production of low yields. This is in direct contrast to the conditions which prevailed in this regard, in the corresponding soil cultures of the preceding experiment, where some of the slightly injured cultures produced relatively high yields and the cultures which suffered pronounced injury gave low yields.

That many of the cultures of group I and group II produced markedly higher yields (relative to the yields from the cultures of the 3-salt group) than did the corresponding cultures of groups III and V, has already been pointed out in connection with a consideration of the dry-weight values. This marked increase in yields was accompanied by a corresponding decrease in the injury sustained by the cultures, as is clearly indicated by the graphs and diagrams of figure 3. Here also it is possible that the antagonistic effects of the new ions, Na and NH_4, introduced into the solutions of group I and group II with NaH_2PO_4 and $(NH_4)H_2PO_4$, respectively, may be responsible for the decrease in the injury produced as well as for the marked improvement in the yields.

5. Relation of acidity to yield and to injury. The H-ion concentration is a characteristic of the culture solutions which may be expected to bear some relation to the yields produced and to the injury sustained by the cultures. It has already been pointed out that the injury sustained by the soybean plants is related to the atomic group H_2PO_4 or to the ions resulting from the partial dissociation of this group in the solutions. The necessary apparatus to determine the H-ion concentration of these solutions electrometrically was not at hand. An attempt was made, however, to determine the acidity of the solutions whose formulas are given in tables 2 and 3, by titration methods, and to relate the results thus obtained to the yield of tops and to the injury sustained by the plants.

In making the acidity determinations, 100 cc. of the solution to be tested was diluted with 150 cc. of distilled water, and was then titrated with $\frac{N}{10}$ NaOH, using as indicators alizarin and methyl orange. Several tests of all the solutions were made with each of these indicators. The results of these tests are

included in table 7, which gives the total acidity (gram-molecules per liter) as here determined, in terms of phosphoric acid. Since the tests made with alizarin yielded results which were in very close agreement with those obtained by the use of methyl orange as an indicator, the results of all the tests for each solution were averaged. The values in the table represent, therefore, the average results of all the tests with each solution.

In order to bring out the relation between the acidity of the solutions and the yields of tops, and also the relation between acidity and injury, the graphs and diagrams of figure 4 were prepared. The dotted line in each set of graphs

TABLE 7

Acidity of culture solutions in terms of phosphoric acid

SOLUTION NUMBER	GRAM MOLECULES PER LITER				
	Series I	Series II	Series III	Series V	3-salt series
$T_1R_1C_1$	0.000199	0.000050	0.000149	0.000497	0.000000
C_3	0.000189	0.000050	0.000149	0.000546	0.000000
C_5	0.000149	0.000050	0.000099	0.000596	0.000000
C_7	0.000174	0.000050	0.000099	0.000546	0.000000
R_3C_1	0.000447	0.000224	0.000274	0.000720	0.000323
C_3	0.000348	0.000199	0.000224	0.000671	0.000298
C_5	0.000398	0.000199	0.000248	0.000546	0.000199
R_5C_1	0.000497	0.000447	0.000472	0.000918	0.000397
C_3	0.000475	0.000447	0.000472	0.000820	0.000397
R_7C_1	0.000621	0.000497	0.000497	0.000992	0.000695
$T_3R_1C_1$	0.000398	0.000199	0.000174	0.001290	0.000050
C_3	0.000298	0.000199	0.000174	0.001240	0.000050
C_5	0.000224	0.000149	0.000174	0.001440	0.000050
R_3C_1	0.000546	0.000348	0.000323	0.001890	0.000397
C_3	0.000447	0.000199	0.000274	0.001740	0.000348
R_5C_1	0.000646	0.000447	0.000447	0.001990	0.000645
$T_5R_1C_1$	0.000646	0.000447	0.000274	0.002640	0.000125
C_3	0.000447	0.000348	0.000224	0.002680	0.000075
R_3C_1	0.000696	0.000645	0.000472	0.003140	0.000497
$T_7R_1C_1$	0.000746	0.000423	0.000497	0.003680	0.000298

represents the actual average yield values and the unbroken line represents the acidity of the solutions (gram-molecules per liter) in terms of phosphoric acid. The values represented by the graphs for each group are here arranged in the descending order of the actual dry-weight values for the 3-salt group. The order of arrangement is indicated by the culture or solution numbers placed below, and is the same as that employed in the preparation of the graphs of figure 3. The relative degrees of injury are again indicated by broad vertical lines, in a manner similar to that employed in the preceding figures.

Inspection of the data of table 7 and the graphs of figure 4, brings out the

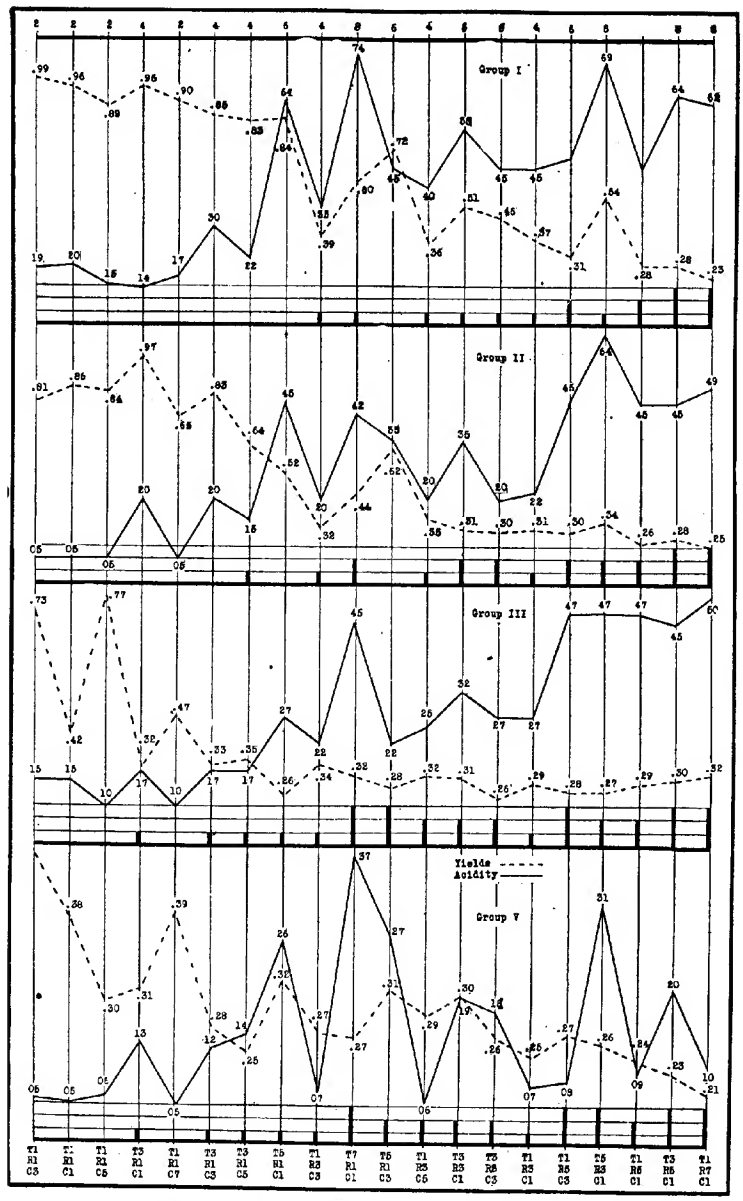

FIG. 4. RELATION OF ACIDITY TO YIELDS AND INJURY: GROUPS I–V. (ACIDITY VALUES ARE INDICATED ON CHART BY OMITTING PREFIX 0.000 FOR GROUPS I, II, AND III, AND 0.00 FOR GROUP V)

fact, as is to be expected, that with the increase in the partial osmotic concentrations of the total phosphates in the solutions, the acidity of the solutions is increased. The increase in acidity, however, is not proportional to the increase in concentration due to phosphates; the graphs, therefore, are somewhat irregular. While the acid content of any one of these solutions is mainly determined by the partial osmotic concentration of the total phosphates, it is modified by the relative proportions of the two phosphates (KH_2PO_4 alone in the solutions of group III) in the solution, and also by the relative proportions of the two constituent salts $Ca(NO_3)_2$ and $MgSO_4$.

It will be observed that while the graphs of figure 4, representing acidity, are very irregular, each graph shows a decided tendency to slope upward to the right. On the other hand, all the graphs representing actual average yields slope downward to the right. This clearly indicates a general tendency for the yields to be low in solutions having high acid content, and high in solutions which have low acid content. It is evident, however, that the relative salt proportions determine the kind of growth (good, medium or poor) which will take place in the solutions with medium or low acid content, since both high and low yields of tops occur with each of the four groups in the solutions thus characterized. When the acid content of the solution becomes excessive, the salt proportions appear to have little influence in determining the kind of growth, since only low yields are produced by the solutions with relatively high acid content. Two exceptions to the last statement are to be noted in cultures $T_5R_1C_1$ and $T_7R_1C_1$ of group I, each of which is high in acid, the former producing a relatively high yield and the latter a medium yield.

The graphs and diagrams of figure 4 indicate clearly that the solutions of each of the four groups here considered which produced no injury, were characterized by having low acidity as compared with that of the solutions which produced injury. Two exceptions to this appear in the case of the two solutions $T_5R_1C_1$ and $T_5R_1C_3$ of each of the groups I and II. Each of these solutions showed relatively high acidity but produced no injury. In group III the four solutions, $T_1R_1C_1$, $T_1R_1C_3$, $T_1R_1C_5$ and $T_1R_1C_7$, which produced no injury, were each characterized by having a lower acid content than any of the solutions of the same group which produced the injury. The uninjured cultures of group V correspond to those of group III, and the solutions of each of these cultures likewise were characterized by having a lower acid content than any solution of the same group which produced injury.

There is a general tendency in each of the four groups here considered for the injury to become more pronounced as the acid content of the solutions increases. This relation is, however, not absolutely definite. It is quite obvious that the relative salt proportions play a very important rôle in either accentuating or diminishing the injury when the acid content of the solutions is sufficiently high to produce the injury. One of two solutions having the same acid content and equal osmotic concentrations, may possess relative salt proportions which are favorable to good growth, while the relative salt propor-

tions of the other may not be well adapted to the growth of the plants. The salt proportions of the former would naturally tend to diminish the injurious effects of the acid, while the tendency of the latter would be to augment the injury. For example, the two solutions, $T_5R_1C_1$ and $T_3R_5C_1$ of group I, possessed the same acid content (0.000646) as this was here determined. The former produced a high yield and suffered no injury, while the latter gave a very low yield and sustained severe jnjury. In group II the four solutions $T_5R_1C_1$, $T_1R_5C_3$, $T_1R_5C_1$ and $T_3R_5C_1$, all had the same acidity (0.000447). The first of these solutions produced a medium yield but suffered no injury, while of the remaining solutions, the first two suffered pronounced injury, the last sustained severe injury, and all three gave very poor yields.

The relation between the injury sustained by the plants and the acid content of the solutions of the 3-salt group, is quite definite, as will appear from a study of figure 5. The data of the 3-salt group are here graphically presented

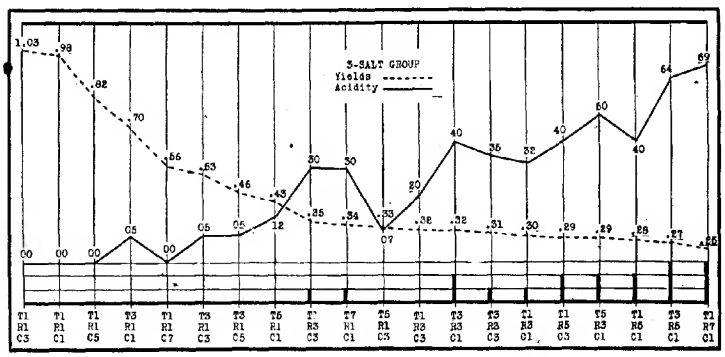

FIG. 5. RELATION OF ACIDITY TO YIELDS AND INJURY: 3-SALT GROUP. (ACIDITY VALUES ARE INDICATED ON CHART BY OMITTING PREFIX 0.000)

in the same manner as are those of the four groups represented in figure 4. It will be observed that each culture of this group with a solution having an acid content higher than 0.000497 showed severe injury. Each culture whose solution had an acidity value of 0.000397 or 0.000497 showed pronounced injury, and each solution with an acid content of 0.000298, 0.000323 or 0.000348 produced slight injury. No injury was produced by any solution of the 3-salt group with an acidity value below 0.000298. A clear relation between acidity and injury is here indicated; the higher the acid content of the solutions above a certain value (0.000298), the greater is the degree of injury sustained by the plants. This relation is definite for the conditions of the present study and for the group of cultures here considered. Hoagland (6) found a somewhat similar relation between the growth of barley seedlings and the H-ion concentration of the medium. An acid condition of the medium was found to be favorable to the growth of the seedlings, and produced no injury when the

acidity was not in excess of a certain limit of the H-ion concentration. When, however, the H-ion concentration was considerably increased beyond the limit set for favorable growth, very decided injury was caused.

It is evident that the injury sustained by the plants of these tests is the result, not of one factor alone, but of combinations of factors. The complexity of the whole problem is such as to render the drawing of definite conclusions extremely difficult. It should be emphasized in this connection that the conclusions here reached with regard to the toxic influence of the various phosphates and salt combinations employed, must be understood to apply for no other set of experimental conditions than those under which these cultures were conducted, and in connection with soybean plants during the early stages of their growth.

SUMMARY

The experiments above presented deal with the influence of five different monobasic phosphates on the growth of soybeans, with special reference to the toxic symptoms produced under various experimental conditions. The tests involved three distinct sets of experiments. (a) The plants were grown in soil cultures to which the salts were added singly in solutions having osmotic concentration values varying from 0.5 to 7 atmospheres. Five groups of cultures corresponding to the five phosphates were employed. Each group comprised 11 cultures including one check culture without salts. (b) The plants were grown in soil cultures to which the phosphates were applied in connection with a complete fertilizer ration in solutions with varying salt proportions but with approximately constant total osmotic concentration values of 2.5 atmospheres. Six groups of cultures (including one control group which employed only the three salts KH_2PO_3, $Ca(NO_3)_2$ and $MgSO_4$) each comprising 20 different sets of salt proportions, were employed. (c) The mixed solutions employed in the preparation of the soil cultures were used also for water cultures, without alteration. A trace of iron as ferric phosphate was added to each solution. For each culture 500 cc. of solution were used.

The total salts added to the soil cultures were included in a single initial application. Each soil culture comprised five plants. With water cultures, the solutions were renewed every third or fourth day and each culture comprised three plants.

The main results of these tests may be summarized as follows:

1. As indicated by relative yields, the growth of soybean tops was injuriously affected by each of the five phosphates employed singly in soil cultures, the average yield from the check cultures being considerably higher than the highest average yield from any of the treated cultures.

2. Each of the phosphates used singly in soil cultures caused specific injury to the plants when the solutions applied to the soil had osmotic concentration values above 1 atmosphere.

3. With respect to their capacities for producing specific injury, the phos-

phates employed singly in the soil cultures arrange themselves in the following order, beginning with the salt which is least toxic: (1) KH_2PO_4, (2) $(NH_4)\ H_2PO_4$, (3) $MgH_4(PO_4)_2$, (4) NaH_2PO_4, (5) $CaH_4(PO_4)_2$.

4. The nature of the injury produced by the five phosphates is identical. The injury is related to the common group H_2PO_4, or to the ions resulting from the dissociation of this group in the soil solution.

5. The five groups of soil cultures each employing a single phosphate in connection with the three salts, KH_2PO_4, $Ca(NO_3)_2$, and $MgSO_4$, produced their average maximum yields in corresponding cultures. The phosphates employed arrange themselves in the following order with respect to the production of maximum yield values, beginning with the highest: (1) $CaH_4(PO_4)_2$, (2) NaH_2PO_4, (3) $(NH_4)H_2PO_4$, (4) KH_2PO_4, and (5) $MgH_4(PO_4)_2$. The maximum yield from the 3-salt group was slightly surpassed only by that from the group employing $CaH_4(PO_4)_2$.

6. The plants of a number of soil cultures from each of the five groups employing a phosphate in connection with a complete fertilizer ration, suffered pronounced injury but with no culture was the injury sufficiently severe to cause the death of the plants before the end of the growth period. With respect to their capacity for producing the injury, the phosphates arrange themselves in the same order as they do when employed singly in soil cultures.

7. No specific injury occurred to the plants in any soil culture of the 3-salt group.

8. In each group of soil cultures except the one employing $CaH_4(PO_4)_2$, specific injury was confined to the cultures prepared with solutions whose partial osmotic concentration values due to total phosphates was not less than six-tenths of the total osmotic concentration. In the group employing $CaH_4(PO_4)_2$, injury was observed in cultures prepared with solutions having four-tenths of their total osmotic concentrations due to total phosphates.

9. The degree of injury sustained by the plants was not determined solely by the total phosphate content of the medium, but was related also to the relative proportions of the constituent salts.

10. With these soil cultures there was no definite relation between injury and yields. In general, pronounced injury was coincident with relatively low yields. Slight injury had no greater tendency to occur with low yields than with high yields.

11. In water cultures, the solutions employing NaH_2PO_4 and those employing $(NH_4)H_2PO_4$ in connection with the three salts KH_2PO_4, $Ca(NO_3)_2$ and $MgSO_4$, produced markedly higher yields with nearly every set of salt proportions, than did the corresponding solutions similarly employing KH_2PO_4 or $MgH_4(PO_4)_2$. This is attributed to the antagonistic effects of the new ions Na and NH_4 introduced into the solutions with NaH_2PO_4 and $(NH_4)H_2PO_4$, respectively.

12. The various phosphates used in water cultures with the 3-salt nutrient solutions (neglecting $CaH_4(PO_4)_2$, which rendered the solutions extremely toxic

to the soybean plants, with each set of salt proportions) arrange themselves in the same order, with respect to the production of maximum yield values, as they do in the corresponding groups of soil cultures. The average maximum yield from the 3-salt group was not equaled by that from any other group of solution cultures.

13. The specific injury sustained by the plants in solution cultures was more pronounced than that sustained by the plants in the corresponding soil cultures. With respect to their relative toxicities when employed in solution cultures with complete nutrient mixtures, the phosphates arrange themselves in the following order, beginning with the salt which is least toxic: (1) NaH_2PO_4, (2) $(NH_4)H_2PO_4$, (3) KH_2PO_4, (4) $MgH_4(PO_4)_2$, and (5) $CaH_4(PO_4)_2$.

14. Careful comparisons of injured plants from all the different media here considered showed the nature of the specific injury sustained by the plants to be identical.

15. Neglecting the solutions employing $CaH_4(PO_4)_2$, no injury was sustained by the plants grown in solutions whose partial osmotic concentrations due to total phosphates was less than four-tenths of the total osmotic concentration.

15. The relation between yields and injury with the solution cultures may be expressed as follows: The greater the degree of injury (considering only injured cultures) the lower the yields. High yields were obtained from solutions which produced no injury.

17. The acid content of the solutions bears a definite relation to the specific injury sustained by the plants grown in them: the higher the acid content (as here determined by titration methods) the greater the degree of injury suffered by the plants. The relative salt proportions, however, play a very important rôle in either accentuating or diminishing the injury when the acidity of the solutions is sufficiently high to produce it.

REFERENCES

(1) Ayres, A. H. 1917 Influence of the composition and concentration of the nutrient solution on plants grown in sand cultures. Univ. Calif. Pub. Agr. Sci., v. 1, no. 11, p. 341–394.

(2) Briggs, L. J., and Shantz, H. L. 1911 A wax seal method for determining the lower limit of available soil moisture. *In* Bot. Gaz., v. 51, p. 210–219.

(3) Gile, P. L. 1912 Lime-magnesium ratio as influenced by concentration. Porto Rico Agr. Exp. Sta. Bul. 12.

(4) Harris, F. S. 1915 Effect of alkali salts in soils on the germination and growth of crops. *In* Jour. Agr. Research, v. 5, no. 1, p. 1–53.

(5) Hilgard, E. W. 1911 Soils, their Formation, Properties and Composition. p. 209. New York.

(6) Hoagland, D. R. 1917 The effect of hydrogen and hydroxyl-ion concentration on the growth of barley seedlings. *In* Soil Sci., v. 3, no. 6, p. 547–560.

(7) Jones, H. C. 1912 The electrical conductivity, dissociation and temperature coefficients of conductivity from zero to sixty-five degrees, of aqueous solutions of a number of salts and organic acids. Carnegie Inst. Washington, Pub. 170.

(8) McCall, A. G. 1916 Physiological balance of nutrient solutions for plants in sand cultures. *In* Soil Sci., v. 2, no. 3, p. 207–253.

(9) McCool, M. M. 1913 The action of certain nutrient and non-nutrient bases on plant growth. N. Y. Agr. Exp. Sta. (Cornell) Mem. 2, p. 121–170.

(10) Osterhout, W. J. V. 1911 The permeability of protoplasm to ions and the theory of antagonism. *In* Science, v. 35, p. 112–115.

(11) Shive, J. W. 1914 The freezing points of Tottingham's nutrient solutions. *In* Plant World, v. 17, no. 12, p. 345–353.

(12) Shive, J. W. 1915 A study of physiological balance in nutrient media. *In* Physiol. Res., v. 1, no. 7, p. 327–395.

(13) Shive, J. W. 1916 The influence of various salts on the growth of soybeans. *In* Soil Sci., v. 1, no. 2, p. 163–170.

(14) Tottingham, W. E. 1914 A quantitative chemical and physiological study of nutrient solutions for plant growth. Physiol. Res., v. 1, no. 4, p. 133–245.

(15) Trelease, S. F. 1917 The relation of concentration of the nutrient solution to the growth of young wheat plants in water cultures. Johns Hopkins Univ. Circ., March, 1917, p. 225–227.

(16) Watkins, C., and Jones, H. C. 1915 Conductivity and dissociation of some rather unusual salts in aqueous solutions. *In* Jour. Amer. Chem. Soc., v. 37, p. 2626–2636.

EFFECT OF AMMONIUM SULFATE IN NUTRIENT SOLUTION ON THE GROWTH OF SOYBEANS IN SAND CULTURES

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INTRODUCTION

Since the middle of the last century a great amount of work has been done in furthering our knowledge on the question of salt requirements for green plants. Perhaps the most logical method of determining the best salt proportions for the growth of plants in water cultures was that recently adopted by Tottingham (29). Using Knop's (13) formula as a basis, Tottingham varied the proportions of the component salts by the increments of one-tenth of the total osmotic concentrations. All possible salt proportions were thus included in his series, necessitating the use of 84 different solutions. Tottingham was thus able to develop a nutrient solution for wheat in its early stage of growth that produced yields 11 per cent higher than did Knop's solution with the same total concentration (osmotic value 2.5 atm.).

Tottingham's solution contains four salts, KH_2PO_4, KNO_3, $Ca(NO_3)_2$ and $MgSO_4$. It will be observed that KNO_3 may be omitted from the Tottingham's solution without omitting any ion which is not furnished by the remaining salts. Thus, by using Tottingham's general method, Shive (26) has found that the omission of KNO_3 from the nutrient solution employed by Tottingham (29) not only did not cause any ill effect on the growth of young wheat plants, but actually produced an increased yield over that from Tottingham's best solution with the same total concentration (osmotic value 1.75 atm.).

This behavior of the 3-salt solution, on the other hand, suggests another possibility. Might not some other salt, although not containing any element that is not already present in the nutrient solution, benefit plant growth if added in proper proportion. Such a salt might be ammonium sulfate. Adding no new elements, it would introduce a new ion that could have some influence on the plant growth when present in different proportions with other ions in the nutrient solution.

It has been demonstrated that some plants can use ammoniacal nitrogen in the absence of NO_3. Gerlach and Vogel (6) grew corn seedlings in sterilized soil with the solution devoid of any nitrates, but containing ammonium sulfate along with the other elements required for plant growth, and found that a considerable amount of nitrogen was absorbed by the plants as compared with the amounts absorbed from untreated soil. Prianishnikov (24) classified the

most important field crops into three groups. The plants of the first group, including corn, barley, pumpkin, etc., can be supported readily by ammonium chloride or ammonium sulfate as a source of nitrogen. The second group (peas, vetches, etc.) suffered injury when treated with ammoniacal nitrogen, while the third group, comprising a single plant, *Lupinis luteus*, can not make a normal growth when treated with this kind of nitrogenous compounds. In connection with the latter plant reference can be made to the work of Nikolaeva (21). Therefore, it is possible that ammonium sulfate may have a nutritive effect in a nutrient solution that may result in the better development of the plants.

It is a well known fact that a cation of any salt, if this salt is present alone in the medium, has a toxic effect on living organisms, while no such injury is observed, if the properly balanced mixture of two or more salts is present in solution. For a very extensive review of literature on this important and interesting subject, reference may be made to McCool's (20) work and also to the exhaustive monograph of Frear (5). Osterhout (22) points out that the injurious effect of the NH_4-ion on plant growth could be antagonized by one of the following ions: Ca, Na and K. McCool's extensive experiments strongly support this contention of Osterhout. Working mainly with chlorides McCool (20) has found that the toxic action of NH_4Cl upon Canada field peas could be overcome, at least in part, by either Ca or Na in the chloride form.

The action of NH_4 in the form of sulfates on the plant tissue, perhaps, differs considerably from that of the same ion in a chloride, but the two cases could well be expected to be in parallel.

In view of the foregoing facts, it was deemed desirable to test this salt with Tottingham's nutrient solution, using the salt to be tested in the place of KNO_3. With our knowledge of the behavior of Tottingham's solution as compared with Shive's (26), an experiment of this nature would throw some light on the effect of ammonium sulfate when added to Shive's nutrient solution.

Two chief methods are employed by plant physiologists in most of the partially controlled experiments, namely, water culture and sand culture methods. Of the two, the sand culture, as a medium, more nearly approaches the natural conditions for plant development, as these conditions exist in soil. This method was chosen for the present work.

EXPERIMENTAL

Method employed

The experimental work herein recorded was done with soybeans of the "Black Eyebrow" variety. The seed came originally from the United States Department of Agriculture and was used on experimental plots at the New Jersey Agricultural Experiment Station.

The plants were grown in sand cultures in earthenware glazed cylindrical

pots that are 12.5 cm. in height having an inside diameter of 10 cm. Fourteen hundred grams of sand, washed in distilled water and dried, was placed in each pot within about 1 cm. of the top. The water-holding capacity of the sand used was 23.40 per cent (the average of two determinations). The moisture content of the medium in which the plants are grown has considerable influence on the plant development, as is well shown by a number of investigators. In this connection the works of Hellriegel (11), Prianishnikov (23), Harris (9), and Tulaikoff (30) are noteworthy. Both Hellriegel and Tulaikoff have found that the best crop yield was obtained with the moisture content of the medium equivalent to about 60 per cent of the water-holding capacity of the soil. In view of these facts it was decided to maintain a proportionally

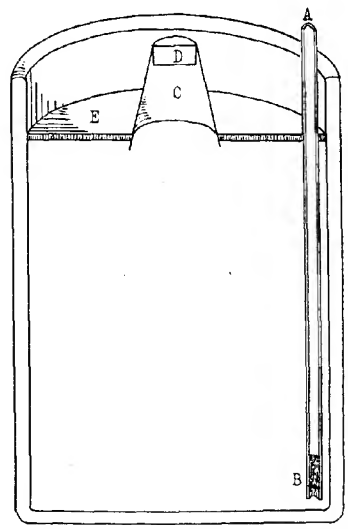

FIG. 1. DIAGRAM OF POT USED IN EXPERIMENTS
(Drawing by Dr. J. W. Shive)

similar moisture content in the sand cultures used in these experiments, which corresponded to 14 per cent of water based on the dry weight of sand in question.

The water content in the cultures was kept as constant as was practicable by adding water to the pots from day to day. For this purpose the pots were weighed every other day, but water was added daily in amount corresponding to its loss during the previous day. Every three or four days the solution in the sand cultures was changed according to a method similar to that employed by McCall (19), the difference being in the design of the pots. Instead of drawing off the solution at the bottom, as was done by McCall, provision was made to withdraw the excess solution at the top by means of a glass tube, which extended to the bottom of the pot. A glance at figure 1 reveals the main

features of the pot used in these experiments. The glass tubing may be of any convenient size and should be somewhat longer than the height of the pot. The lower portion was filled with glass wool to serve as a filter for the fine particles of sand. A small rubber tube was attached to the upper end

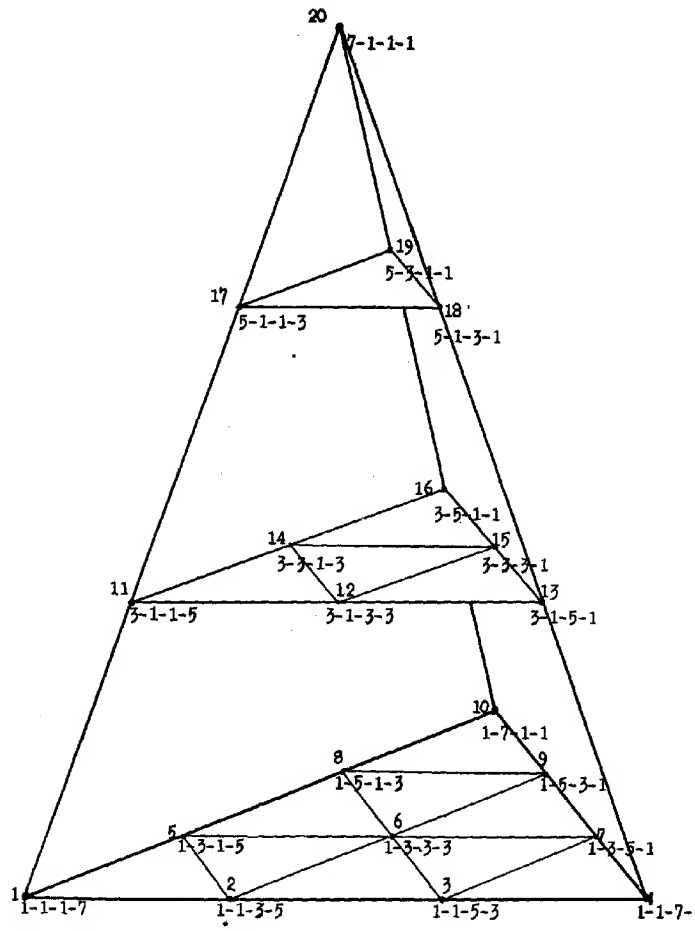

FIG. 2. SKELETON OF A PYRAMID SHOWING THE REPRESENTATIVE CULTURES USED IN THE PRESENT WORK

of the glass tube and thus served for connecting the pot with the suction outfit, which had a graduated cylinder to catch the solution. The changing of the solution was effected in the same manner as described by McCall (19). After planting the seedlings the surface of the sand in the pots was sealed

with a mixture of parowax and vaseline prepared according to the formula of Briggs and Schantz. The water and solution were added through the paper funnel. The paper funnels used in these experiments were prepared by rolling the manilla paper to form a cone, which was dipped into melted paraffine and then cooled. This was done to make it hold together. It was then cut to the desired length, inserted into the sand medium about 2 cm. deep in the middle of the pot, and corked. The length of the funnel used was 6 to 7 cm.

TABLE 1

Osmotic requirements of cultures

The figures in the table represent the number of cubic centimeters of the given salts of m/4 concentrations needed, when mixed together, and diluted to 250 cc., to make the concentration of resultant solution equal to approximately 2.5 atmospheres of pressure

POSITION OF CULTURES IN THE TRIANGLES OF THE PYRAMID	TOTTINGHAM SERIES					AMMONIUM SULFATE SERIES				
	Number of cultures	KH_2PO_4 m/4	KNO_3 m/4	$Ca(NO_3)_2$ m/4	$MgSO_4$ m/4	Number of cultures	KH_2PO_4 m/4	$(NH_4)_2SO_4$ m/4	$Ca(NO_3)_2$ m/4	$MgSO_4$ m/4
$T_1R_1C_1$	1	5.2	4.9	3.6	40.6	21	5.2	3.6	3.6	40.6
C_3	2	5.2	4.9	10.8	29.0	22	5.2	3.6	10.8	29.0
C_5	3	5.2	4.9	18.0	17.4	23	5.2	3.6	18.0	11.6
C_7	4	5.2	4.9	25.2	5.8	24	5.2	3.6	25.2	5.8
R_3C_1	5	5.2	14.6	3.6	29.0	25	5.2	10.8	3.6	29.0
C_3	6	5.2	14.6	10.8	17.4	26	5.2	10.8	10.8	17.4
C_5	7	5.2	14.6	18.0	5.8	27	5.2	10.8	18.0	5.8
R_5C_1	8	5.2	24.4	3.6	17.4	28	5.2	18.0	3.6	17.4
C_3	9	5.2	24.4	10.8	5.8	29	5.2	18.0	10.8	5.8
R_7C_1	10	5.2	34.2	3.6	5.8	30	5.2	25.2	3.6	5.8
$T_3R_1C_1$	11	15.6	4.9	3.6	29.0	31	15.6	3.6	3.6	29.0
C_3	12	15.6	4.9	10.8	17.4	32	15.6	3.6	10.8	17.4
C_5	13	15.6	4.9	18.0	5.8	33	15.6	3.6	18.0	5.8
R_3C_1	14	15.6	14.6	3.6	17.4	34	15.6	10.8	3.6	17.4
C_3	15	15.6	14.6	10.8	5.8	35	15.6	10.8	10.8	5.8
R_5C_1	16	15.6	24.4	3.6	5.8	36	15.6	18.0	3.6	5.8
$T_5R_1C_1$	17	26.0	4.9	3.6	17.4	37	26.0	3.6	3.6	17.4
C_3	18	26.0	4.9	10.8	5.8	38	26.0	3.6	10.8	5.8
R_3C_1	19	26.0	14.6	3.6	5.8	39	26.0	10.8	3.6	5.8
$T_7R_1C_1$	20	36.4	4.9	3.6	5.8	40	36.4	3.6	3.6	5.8

Solutions used

The solutions employed in this work were the same as in the work of Tottingham (29), who varied the osmotic concentration of his salt solution by one-tenth of their total osmotic concentration. The entire set of 84 cultures, however, was not used, but only 20 of the most representative cultures were selected, the selection being done in the following manner. The second, fourth and sixth triangles were entirely omitted. In the remaining first, third and fifth triangles every other row was omitted, and every other culture was

omitted in the remaining rows. This arrangement leaves 19 cultures in the three mentioned triangles. The twentieth culture used corresponded to the apex of the pyramid. Figure 2 gives a general idea of the place in the pyramid of the cultures employed. It will be seen to be a fairly representative skeleton. It also considerably lessens the work.

In parallel with the Tottingham cultures, another series of cultures was used, in which, instead of KH_2PO_4, KNO_3, $Ca(NO_3)_2$ and $MgSO_4$, the following salts were used: KH_2PO_4, $(NH_4)_2SO_4$, $Ca(NO_3)_2$ and $MgSO_4$, or the place of KNO_3 was taken by $(NH_4)_2SO_4$, thus partially eliminating the potassium and nitrogen, and introducing an entirely new ion, NH_4. The purpose of this substitution, as was stated in the introduction, was to find out whether or not a new ionic group, which adds no new element to the culture solution, might influence the crop yield in the sand cultures. Iron was supplied in the form of a small amount of iron rust.

The total osmotic concentration of the nutrient solutions used was about 2.5 atmospheres in terms of possible osmotic pressure. The calculations for volume-molecular concentration of ammonium sulfate were made according to the method adopted by Tottingham, using the dissociation figures from Jones' tables (12). The stock solutions were prepared in M/4 concentration. The needed amount of solution of the different salts was drawn into a small volume of distilled water in a 250-cc. volumetric flask which was then filled to the mark. The number of cubic centimeters of different stock solutions necessary for obtaining the nutrient solution with the osmotic concentration of approximately 2.5 atmospheres is given for convenience in table 1.

The plants were started in moist sand, and, when about 2 cm. in height, were selected for uniformity and transplanted into the culture pots. Three healthy seedlings were used in each pot. After transplanting, the surface of the sand, as mentioned above, was sealed with paraffine.

Forty sand cultures, twenty in each of the two series thus prepared, were set on January 25, 1917, and continued to March 5, the total growing period in pots being 39 days. Later the experiment was repeated, and was run from April 2 to May 11 of the same year, again making 39 days for the growing season. The plants, when harvested, were in full bloom, while on the most vigorous ones the pods were being formed.

RESULTS AND DISCUSSION

Concentration of the nutrient solution

In order to ascertain whether an appreciable adsorption of salts had taken place after adding the solutions of these salts to the sand, the concentration of the solutions was determined both before and after adding them to the sand. Moreover, the concentration of the solutions in plant cultures also was deter-

mined. This was done at three different times during the growth: (a) at the beginning of the experiment, (b) 15 days after the start, and (c) at the end of the 39-day period of growth. A portion of the solution was withdrawn by means of suction from each of the sand cultures. This was done at the time of renewing the solutions. In addition to this, the concentration of the moist sand at the end of the experiment also was determined by means of the cryoscopic method. The concentration of the solution in the sand was determined by the method suggested by Bouyoucos and McCool (2), and the calculations were made with the aid of data gathered by Harris and Gortner (10).

Table 2 gives the data obtained by these determinations with the Tottingham series while table 3 presents the similar data for the ammonium sulfate series. The arrangement of the data is the same in both tables. The first two columns represent the number of cultures in the triangles and the laboratory numbers, respectively. The following seven columns give the degrees (Centigrade) of depression of the freezing point, while the last seven columns show the corresponding concentrations in atmospheres. Studying these tables carefully one notices two very significant features. One of these lies in the fact that, with few exceptions, the values for the original solution in both series, for the sand treated with solution, for all solutions obtained by suction from the culture pots, and, finally, for the sand at the end of the experiment, follow one another with striking consistency. There is no great difference between the concentration of the original solution and that of the solution in the sand cultures. In the Tottingham series the values for solutions obtained from the sand cultures are in very close agreement with the values of the original solution and sand treated with it. The figures for the sand at the end of the experiment, however, are lower than those of others, but this latter feature is not so noticeable in the series containing ammonium sulfate. Thus, it is evident that Livingston's (15) results, as emphatically quoted by Breazeale (3), could not be explained on the basis of total concentration. His results seem to indicate that while the best concentration of solution in water cultures for plant growth is around 300 parts per million, the corresponding best concentration in sand cultures lies in the neighborhood of 2500 parts per million. Unless very fine sand is used and the most of the salts composing the solution are similar in their behavior to phosphates, no such reduction in the effective concentration is to be expected. In this connection the work of Bouyoucos and McCool (2) with a number of soils and sand which they treated with different salts of $\frac{N}{10}$ concentration, is of special interest. Testing the total concentration directly in the soil treated with the salt solutions, Bouyoucos and McCool obtained results which show that different salts are adsorbed to a different degree by the same soil, and that the different soils and sands have different powers of adsorption. The adsorption of sand, which they treated with $\frac{N}{10}$ salt solution, was very small. However, when the solution of a salt is sufficiently diluted, the relative adsorption of even coarse sand becomes considerable and that of the fine sand adsorption is very great, as was shown by

TABLE 2

Freezing-point depression and the concentration in atmospheres of Tottingham's solutions

POSITION	NUMBER	FREEZING-POINT DEPRESSION							CONCENTRATION IN ATMOSPHERES						
		Original solutions	Sand treated with solutions	Difference	Solution obtained by suction from pots at the beginning of experiment	Solution obtained by suction from pots 15 days after beginning	Solution obtained by suction at the end of the experiment	Sand in pots at the end of the experiment	Original solution	Sand treated with original solution	Difference	Solution obtained by suction from pots at the beginning of the experiment	Solution obtained by suction from pots 15 days after beginning of experiment	Solution obtained by suction from pots at the end of the experiment	Sand in pots at the end of the experiment
		°C.	°C.	°C.	°C.	°C.	°C.	°C	*atm.*	*atm.*	*atm.*	*atm.*	*atm.*	*atm.*	*atm.*
$T_1R_1C_1$	1	0.169	0.154	0.015	0.161	0.185	0.159	0.111	2.04	1.86	0.18	1.94	2.23	1.92	1.34
R_1C_3	2	0.180	0.178	0.002	0.173	0.197	0.166	0.150	2.17	2.15	0.02	2.09	2.38	2.00	1.81
R_1C_5	3	0.184	0.169	0.015	0.180	0.173	0.173	0.148	2.22	2.04	0.18	2.17	2.09	2.09	1.79
R_1C_7	4	0.208	0.205	0.003	0.200	0.201	0.191	0.178	2.51	2.47	0.04	2.41	2.42	2.30	2.15
R_3C_1	5	0.179	0.155	0.024	0.168	0.185	0.165	0.146	2.16	1.87	0.29	2.03	2.23	1.99	1.76
R_3C_3	6	0.172	0.145	0.027	0.162	0.173	0.167	0.155	2.07	1.75	0.32	1.95	2.09	2.01	1.87
R_3C_5	7	0.201	0.201	0.000	0.194	0.203	0.188	0.175	2.42	2.42	0.00	2.34	2.44	2.27	2.11
R_5C_1	8	0.183	0.171	0.012	0.163	0.163	0.158	0.144	2.21	2.06	0.15	1.97	1.97	1.91	1.74
R_5C_3	9	0.208	0.198	0.010	0.174	0.198	0.181	0.155	2.51	2.39	0.12	2.10	2.39	2.18	1.87
R_7C_1	10	0.201	0.197	0.004	0.171	0.193	0.164	0.151	2.42	2.38	0.04	2.06	2.33	1.98	1.82
$T_3R_1C_1$	11	0.176	0.172	0.004	0.182	0.190	0.166	0.146	2.12	2.07	0.05	2.20	2.29	2.00	1.76
R_1C_3	12	0.173	0.165	0.008	0.179	0.195	0.170	0.150	2.09	1.99	0.10	2.16	2.35	2.05	1.81
R_1C_5	13	0.206	0.199	0.007	0.188	0.199	0.180	0.180	2.48	2.40	0.08	2.27	2.40	2.17	2.17
R_3C_1	14	0.171	0.162	0.009	0.185	0.178	0.160	0.130	2.06	1.95	0.11	2.22	2.15	1.93	1.46
R_3C_3	15	0.195	0.193	0.002	0.198	0.196	0.184	0.160	2.35	2.33	0.02	2.39	2.36	2.22	1.93
R_5C_1	16	0.192	0.191	0.001	0.196	0.185	0.181	0.216	2.32	2.30	0.02	2.36	2.23	2.18	2.60
$T_5R_1C_1$	17	0.175	0.169	0.006	0.166	0.164	0.157	0.161	2.11	2.04	0.07	2.00	1.98	1.89	1.94
R_1C_3	18	0.199	0.187	0.012	0.193	0.200	0.182	0.174	2.40	2.26	0.14	2.33	2.41	2.20	2.10
R_3C_1	19	0.189	0.190	−0.001	0.199	0.194	0.188	0.184	2.28	2.29	−0.01	2.40	2.34	2.27	2.22
$T_7R_1C_1$	20	0.200	0.199	0.001	0.194	0.200	0.181	0.175	2.41	2.40	0.01	2.34	2.41	2.18	2.11
Average		0.188	0.180	0.008	0.181	0.189	0.173	0.161	2.27	2.17	0.10	2.18	2.28	2.09	1.94

Freezing-point depression and concentration in atmospheres of solutions of the ammonium sulfate series

POSITION	NUMBER	FREEZING-POINT DEPRESSION[1]							CONCENTRATION IN ATMOSPHERES						
		Original solutions	Sand treated with solutions	Difference	Solution obtained by suction from pots at the beginning of experiment	Solution obtained by suction from pots 15 days after beginning	Solution obtained by suction from pots at the end of experiment	Sand in pots at the end of the experiment	Original solution	Sand treated with original solution	Difference	Solution obtained by suction from pots at the beginning of experiment	Solution obtained by suction from pots 15 days after the beginning of experiment	Solution obtained by suction from pots at the end of experiment	Sand in pots at the end of experiment
		°C.	°C.	°C.	°C.	°C.	°C.	°C.	atm.	atm.	atm.	atm.	atm.	atm.	atm.
$T_1R_1C_1$	21	0.169	0.183	−0.014	0.174	0.196	0.168	0.201	2.04	2.24	−0.17	2.10	2.36	2.03	2.42
C_3	22	0.179	0.186	−0.007	0.190	0.205	0.186	0.223	2.16	2.24	−0.08	2.47	2.47	2.24	2.69
C_5	23	0.179	0.201	−0.022	0.188	0.175	0.182	0.172	2.16	2.42	−0.26	2 27	2.10	2.20	2.07
C_7	24	0.204	0.224	−0.020	0.198	0.225	0.203	0.164	2.46	2.70	−0.24	2.39	3.10	2.45	1.98
R_3C_1	25	0.167	0.205	−0.038	0.174	0.185	0.158	0.195	2.01	2.47	−0.46	2.10	2.23	1.91	2.35
C_3	26	0.174	0.202	−0.028	0.176	0.190	0.170	0.166	2.10	2.44	−0.31	2.12	2.29	2.05	2.00
C_5	27	0.196	0.204	−0.008	0.202	0.215	0.195	0.199	2.36	2.46	−0.10	2.44	2.59	2.35	2.40
R_5C_1	28	0.168	0.168	0.000	0.165	0.178	0.163	0.162	2.03	2.03	0.00	1.99	2.15	1.97	1.95
C_3	29	0.188	0.205	−0.017	0.198	0.197	0.191	0.215	2.27	2.47	−0.20	2.39	2.38	2.30	2.59
R_7C_1	30	0.167	0.184	−0.017	0.189	0.200	0.183	0.184	2.01	2.22	−0.21	2.28	2.41	2.21	2.22
$T_3R_1C_1$	31	0.174	0.193	−0.019	0.185	0.197	0.174	0.129	2.10	2.33	−0.23	2.23	2.38	2.10	1.45
C_3	32	0.174	0.179	−0.005	0.182	0.198	0.180	0.162	2.10	2.16	−0.06	2.20	2.39	2.17	1.95
C_5	33	0.198	0.205	−0.007	0.199	0.230	0.198	0.211	2.39	2.47	−0.08	2.40	2.77	2.39	2.54
R_3C_1	34	0.173	0.183	−0.010	0.176	0.192	0.166	0.191	2.09	2.21	−0.12	2.12	2.32	2.00	2.30
C_3	35	0.201	0.221	−0.020	0.178	0.208	0.194	0.224	2.42	2.66	−0.16	2.15	2.51	2.34	2.70
R_5C_1	36	0.200	0.191	0.009	0.182	0.208	0.182	0.231	2.41	2.30	0.11	2.20	2.51	2.20	2.21
$T_5R_1C_1$	37	0.158	0.148	0.010	0.155	0.211	0.171	0.183	1.91	1.79	0.12	1.87	2.54	2.06	2.21
C_3	38	0.194	0.176	0.018	0.187		0.192	0.201	2.34	2.12	0.22	2.25		2.32	2.42
R_3C_1	39	0.181	0.184	−0.003	0.190	0.197	0.180	0.214	2.18	2.22	−0.04	2.29	2.38	2.17	2.58
$T_7R_1C_1$	40	0.193	0.184	0.009	0.186	0.214	0.192	0.200	2.33	2.22	0.11	2.24	2.58	2.32	2.41
Average		0.177	0.191	−0.014	0.184	0.200	0.176	0.176	2.13	2.30	−0.17	2.22	2.41	2.12	2.12

Wolkoff (33) in the preliminary paper on the study of adsorption of ammonium sulfate at different concentrations.

Another outstanding feature of the results submitted in tables 2 and 3 and graphically presented in figure 3, is the apparent tendency of the nutrient solution containing ammonium sulfate to become more concentrated than the original solution, when added to the sand. In fifteen cases out of twenty the freezing-point depression of the solution in sand was greater than that of the corresponding original solution. The behavior of the Tottingham solution was entirely different in this respect. In nineteen cases out of twenty the concentration of the solution in sand was somewhat smaller than the concentration of the solution before adding this sand. The solutions used in this particular experiment were prepared together and used at once. The same stock solutions were employed for the determination of concentration of the

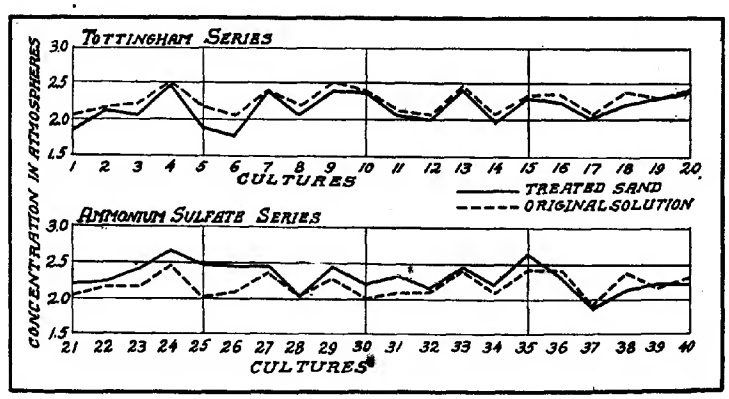

FIG. 3. THE CONCENTRATION OF TOTTINGHAM NUTRIENT SOLUTION AND SOLUTION OF AMMONIUM SULFATE SERIES BEFORE AND AFTER ADDING THESE SOLUTIONS TO A WASHED SEA SAND

solution as such and when added to the sand. Duplicates, of course, were made in every instance. Besides, in the cases where the differences between the original solutions and the solutions in sand were very pronounced, the experiment was repeated. No evident error was revealed. These results are in good agreement with the data obtained by the author (33) working with ammonium sulfate solutions and sand of different fineness. Evidently, when ammonium sulfate is present in combination with other salts, it has a similar effect on the total concentration of the solution as it has when used singly, although the degree of the effect produced is modified by the presence of other salts and also by the salt proportions. The explanation for this interesting phenomenon is offered by the author in another paper (33). It seems that when sand or soil is treated with ammonium sulfate, the alkali radical, NH_4, is adsorbed by this solid phase more strongly than the acid radical, SO_4. As a

result of this unequal adsorption some excess of the SO_4-ion is left in solution, and, uniting with H_2-ions of slightly dissociated water, forms some free H_2SO_4. Since the dissociation of acid is greater than that of the original salt, ammonium sulfate, the freezing-point depression in the case of sand will be greater, although the total (absolute) concentration of the solution may be slightly reduced by the adsorption. This phenomenon is more pronounced in the sand with a lower per cent of moisture than with a higher one. In other words, where the solid phase is relatively reduced and the relative surface is correspondingly diminished, the effect of adsorption becomes smaller. This fact, of course, is in accord with the theory of adsorption. In the case of soil (33) treated with ammonium sulfate, however, no such apparent increase in total concentration was noticed by this method, because the effect of adsorption is greater than that of any increase in dissociation due to the formation of acid.

This phenomenon of increase in concentration of the salt solution on adding it to the sand is similar to that which was first observed by Graham (8) in 1830, but attributed by him to experimental error. The fact of increase of concentration of some salt solutions after adding them to some solid phase, such as charcoal, kaolin, clay, or powdered sand, is now well established by Gore (7), Lagergren (14), William (32) and quite recently by McCall and his co-workers (16). Dealing with the solid phase greatly divided to offer an enormous surface energy, the above-mentioned authors evidently had the adsorption of water out of the liquid phase to such an extent that it caused an increase in concentration of the remaining salt solution. The phenomenon is called either "negative adsorption" (Lagergren, William), or "special case of selective adsorption" (McCall). In the experiments described in the present work, the phenomena observed were evidently due to a selective adsorption of one of the ions and subsequent formation of acid out of the acid radical, and the hydrogen ion of the slightly ionized water. It is a pure case of a physico-chemical process that may take place in the heterogenous system.

RESULTS OF THE MAIN EXPERIMENT

Introductory remarks

The effect of the introduction of ammonium sulfate into Shive's nutrient solution, or of the substitution of this salt for potassium nitrate in Tottingham's nutrient solution, on the plant growth of soybeans was studied by means of several criteria; (a) the general appearance of plants, (b) dry weight of tops, (c) dry weight of roots, (d) transpiration, (e) water requirement of tops, and (f) water requirement of roots.

General appearance of plants of the two series

The plants of the ammonium sulfate series appeared, in general, more healthy, more bushy and slightly taller, than the plants in the corresponding cultures of the Tottingham series. The most pronounced difference was in

the color of the plants of the two series, the plants of the ammonium sulfate series being much greener than those of the Tottingham series.

At the beginning of the growing period, plants with ammonium sulfate as a part of their nutrient solution appeared to be injured and were retarded in their growth for a week or ten days. This feature, however, gradually disappeared, and the plants began to make a vigorous growth.

Just before harvesting the plants of the first experiment, on March 5, observations on the cultures were made, noting the injury, if any, and the degree

TABLE 4

Injury of plants in Tottingham and ammonium sulfate series of the first experiment (the observations were made just before harvesting the plants.)

TOTTINGHAM SERIES	AMMONIUM SULFATE SERIES
1. Two plants slightly injured	21. No injury
2. No injury	22. No injury
3. No injury	23. No injury
4. No injury	24. No injury
5. Severe injury of all plants	25. Very slight injury
6. Two plants slightly injured	26. No injury
7. Two plants slightly injured	27. No injury
8. Severe injury of all plants	28. Considerable injury, all plants stunted
9. Apparent injury slight, but plants stunted	29. No injury
10. Severe injury, plants stunted	30. Considerable injury; all plants stunted
11. No injury	31. No injury
12. No injury	32. No injury
13. Plants conspicuously yellow but no apparent injury noticed	33. No injury
	34. One plant died from mechanical injury; otherwise, no injury
14. Severe injury	
15. Slight injury	35. No injury
16. Severe injury	36. Very slight injury
17. Slight injury	37. No injury
18. Slight injury	38. Four leaves of one plant wilted around the margins
19. Severe injury	
20. Severe injury; leaf edges wilted, as if burned	39. No injury
	40. No injury

of the injury. Each culture was judged as a whole. Brief memoranda are given in table 4.

A glance at the memoranda given in table 4 reveals the fact that, with the exception of cultures 28 and 40, the plants of the ammonium sulfate series suffered much less injury than the corresponding ones of the Tottingham series.

In the second experiment, which was conducted between April 2 and May 11, these differences were more pronounced; that is to say, the difference between the best culture in the ammonium sulfate series and the best culture of the Tottingham series in the second experiment was much greater than in the first experiment. On the other hand, the difference between the poorest cul-

tures of the two series in both experiments was also more pronounced during the second experiment. The injury of the plants of cultures 28 and 30 in the second experiment was such that nearly all plants died in the course of two weeks; while the luxuriant growth of culture 32 was more conspicuous in comparison with the growth of the plants in the corresponding Tottingham's culture, No. 12. The difference in the behavior of the plants grown at different times of the year is attributed, at least partly, to the difference in the temperature of the greenhouse. This temperature, on the average, was much higher during the second experiment than during the first one. Since all the chemical reactions proceed with a greater speed at an elevated temperature than at a lower one, the difference in the effect of the salts in different proportions at different temperatures could easily be explained on these grounds.

Outside of this feature, the general trend of the growth of plants in both experiments was practically the same, as one will notice in studying the data of the dry weight of tops and roots in both series which will be shortly presented.

Dry weight of tops

The dry weight of tops was obtained by cutting the plants at the surface of the sand, and drying them in weighed, wide-mouthed bottles in the electric oven at about 108°C. until constant weight was attained. The results are given in table 5. They show the dry weight of tops secured in the first and the second experiments, and the average for the two. The relative values also are given. The first four columns of the table present the data of the Tottingham series, while the last four present the corresponding values for the ammonium sulfate series. The relative values were obtained by dividing each number by the average dry weight of the two series of both experiments, this average figure being 2.3881 gm. These relative values are plotted in figure 4, following the general scheme (with minor modifications) of representation as suggested by Tottingham (29) and also by Shive (26). This chart shows the cultures in their proper places on the triangles. The cultures whose relative values are 110 or above, are shaded by small crosses, while the portion of the triangles with cultures of values of 90 or lower are shaded by small circles. The examination of the average data in table 5 and figure 4 show striking differences between the corresponding values of the cultures in the two series. The best yields in the Tottingham series were obtained from cultures $T_3R_1C_1$ and $T_3R_1C_3$ with the salt proportions corresponding to 0.0156 m. KH_2PO_4, 0.0049 m. KNO_3, 0.0036 m. Ca $(NO_3)_2$, and 0.00290 m. $MgSO_4$ in the first case, and 0.0156 m. KH_2PO_4, 0.0049 m. KNO_3, 0.0108 m. $Ca(NO_3)_2$ and 0.0174 m. $MgSO_4$ in the second case. The best culture of the ammonium sulfate series was $T_3R_1C_1$, or that which contained 0.0156 m. KH_2PO_4, 0.0036 m. $(NH_4)_2SO_4$, 0.0108 m. $Ca(NO_3)_2$, and 0.0174 m. $MgSO_4$. The poorest culture in both series was $T_1R_7C_1$. It contained the greatest amount of KNO_3 in the Tottingham series and the corresponding amount of $(NH_4)_2SO_4$ in the

ammonium sulfate series. The best culture of the ammonium sulfate series, however, was considerably better than the best culture of the Tottingham series, while of the poorest cultures of the two series the culture of Tottingham's solution was better. The variation between the best and the poorest cultures was much greater in the ammonium sulfate series than in the Tottingham series.

The plant growth of the cultures containing ammonium sulfate, was much

TABLE 5

Dry weight of tops of soybeans grown in sand cultures of the Tottingham and ammonium sulfate series

POSITION	TOTTINGHAM SERIES					AMMONIUM SULFATE SERIES				
	Number	Trial 1	Trial 2	Average	Relative value	Number	Trial 1	Trial 2	Average	Relative value
		grams	*grams*	*grams*			*grams*	*grams*	*grams*	
$T_1R_1C_1$	1	2.3390	2.6975	2.5183	106	21	2.2330	3.8747	3.0539	126
R_1C_3	2	2.4716	2.5170	2.4942	101	22	2.6100	4.9817	3.7959	156
R_1C_5	3	2.3340	2.1323	2.2332	95	23	2.5575	4.7962	3.6769	151
R_1C_7	4	2.3322	1.6865	2.0094	86	24	2.6150	4.7812	3.6981	152
R_3C_1	5	1.7714	2.4997	2.1356	89	25	1.4833	2.2685	1.8759	78
R_3C_3	6	2.4210	2.0950	2.2580	96	26	2.0996	4.2710	3.1853	131
R_3C_5	7	2.0846	1.6547	1.8697	80	27	2.1760	3.6059	2.8910	119
R_5C_1	8	1.8292	2.1405	1.9849	83	28	1.4807	0.3798	0.9303	42
R_5C_3	9	1.9480	1.3702	1.6591	72	29	1.8512	2.7548	2.3030	96
R_7C_1	10	1.3316	1.6507	1.6412	63	30	1.2014	0.5235	0.8624	37
$T_3R_1C_1$	11	2.5590	2.7285	2.6438	112	31	2.7491	3.5485	3.1488	132
R_1C_3	12	2.7210	2.4598	2.5904	112	32	3.1743	5.2335	4.2039	173
R_1C_5	13	1.8848	1.5540	1.7174	74	33	2.5979	3.4932	3.0456	127
R_3C_1	14	2.1020	2.1152	2.1086	90	34	2.0410	2.6805	2.3608	99
R_3C_3	15	1.9934	2.0113	2.0044	85	35	2.2241	3.7773	3.0007	124
R_5C_1	16	1.7570	2.0312	1.8941	80	36	1.3306	1.3877	1.3592	58
$T_5R_1C_1$	17	2.4636	2.1007	2.2822	97	37	2.6422	3.2525	2.9474	124
R_1C_3	18	2.2678	1.7015	1.9847	86	38	2.5464	2.8220	2.6843	114
R_3C_1	19	1.8850	2.2182	2.0516	87	39	1.9795	2.0187	1.9991	85
$T_7R_1C_1$	20	2.2200	2.5140	2.3670	100	40	2.1805	2.6370	2.4088	101
Average					89.5	Average				111

better, on the whole, than that of the cultures without this salt. This fact is very well illustrated by figure 4. Indeed, only two cultures in the Tottingham series gave values above 110, while the large shaded portion with crosses on the ammonium sulfate side shows that twelve out of a total of twenty cultures exceeded this value. On the other hand, while twelve cultures of the Tottingham series fell on or below 90, only five cultures of the ammonium sulfate series were found below this value. Consequently, very few cultures were located between the high and the low areas in the case of the ammonium

sulfate series, as compared with the Tottingham series. There is a pronounced tendency for the cultures of the ammonium sulfate series to show considerable

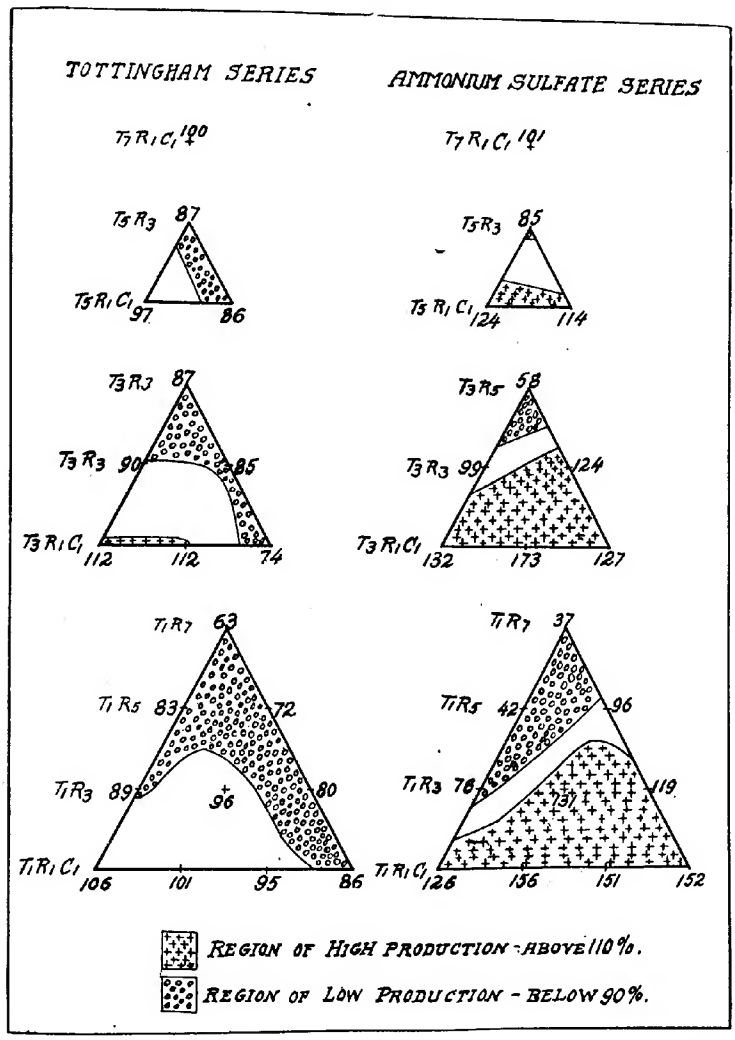

FIG. 4. THE RELATIVE AVERAGE VALUES FOR TOP YIELDS OF SOYBEANS OF TOTTINGHAM AND AMMONIUM SULFATE SERIES

The high and low yield regions are shaded with crosses and circles, respectively.

fluctuation upon a slight alteration of the salt proportions. The fluctuations in this respect are not so striking in the Tottingham series.

Dry weight of roots

The dry weight of roots was obtained by washing out the sand from the roots by means of a small stream of water on a fine screen and drying the roots in the electric oven. Since it is an impossible task to wash the sand from the roots, the weight of organic matter of the roots was obtained in an indirect way. The washed roots were burned in a platinum dish. The difference between the weight of the dish with the material before and after the

TABLE 6

Dry weight of roots of soybeans grown in sand cultures of the Tottingham and ammonium sulfate series

POSITION	TOTTINGHAM SERIES					AMMONIUM SULFATE SERIES				
	Number	Trial 1	Trial 2	Average	Relative value	Number	Trial 1	Trial 2	Average	Relative value
		gram	*gram*				*gram*	*gram*		
$T_1R_1C_1$	1	0.4000	0.4210	0.4105	108	21	0.4590	0.4334	0.4462	117
R_1C_3	2	0.4092	0.4403	0.4248	112	22	0.5868			
R_1C_5	3	0.4536	0.4335	0.4436	116	23	0.4902	0.7275	0.6089	160
R_1C_7	4	0.4436	0.3800	0.4118	108	24	0.3630	0.6405	0.5018	182
R_3C_1	5	0.3132	0.4057	0.3595	95	25	0.2874	0.2305	0.2590	68
R_3C_3	6	0.4628	0.2068	0.3348	88	26	0.4392	0.5745	0.5069	133
R_3C_5	7	0.3472	0.2553	0.3013	79	27	0.3314	0.4101	0.3708	98
R_5C_1	8	0.3118	0.3981	0.3550	94	28	0.2190	0.0398	0.1294	34
R_5C_3	9	0.3230	0.2667	0.2999	78	29	0.2824	0.2322	0.2573	63
R_7C_1	10	0.2296	0.3952	0.3124	82	30	0.1436	0.0410	0.0923	25
$T_3R_1C_1$	11	0.3950	0.4510	0.4230	112	31	0.5118	0.4283	0.4701	124
R_1C_3	12	0.4728	0.4443	0.4586	121	32	0.5616	0.7948	0.6782	179
R_1C_5	13	0.3620	0.3795	0.3708	98	33	0.4864	0.4680	0.4772	125
R_3C_1	14	0.3700	0.3857	0.3779	99	34	0.2500	0.3365	0.2933	78
R_3C_3	15	0.3638	0.3428	0.3533	93	35	0.4350	0.4525	0.4438	117
R_5C_1	16	0.2838	0.3805	0.3322	88	36	0.2132	0.1393	0.1763	47
$T_5R_1C_1$	17	0.4850	0.3575	0.4213	111	37	0.4676	0.4252	0.4464	118
R_1C_3	18	0.4130	0.2960	0.3545	93	38	0.4422	0.4133	0.4278	113
R_3C_1	19	0.3324	0.4483	0.3904	103	39	0.2894	0.2108	0.2501	66
$T_7R_1C_1$	20	0.3718	0.4105	0.3912	103	40	0.4926	0.2873	0.3900	103
Average					99	Average				101

burning was taken as the weight of the roots. No corrections were made for the ash content.

The dry weight of roots, as presented in table 6 and figure 5, shows, in general, the same main differences between the two series, as were pointed out in the case of the tops.

The culture producing the best root yield in the Tottingham series was $T_3R_1C_3$, or one of the two best cultures for tops of the same series. It was

the best culture for roots and tops in the ammonium sulfate series. The poorest culture for roots in the ammonium sulfate series was $T_1R_7C_1$, or the same

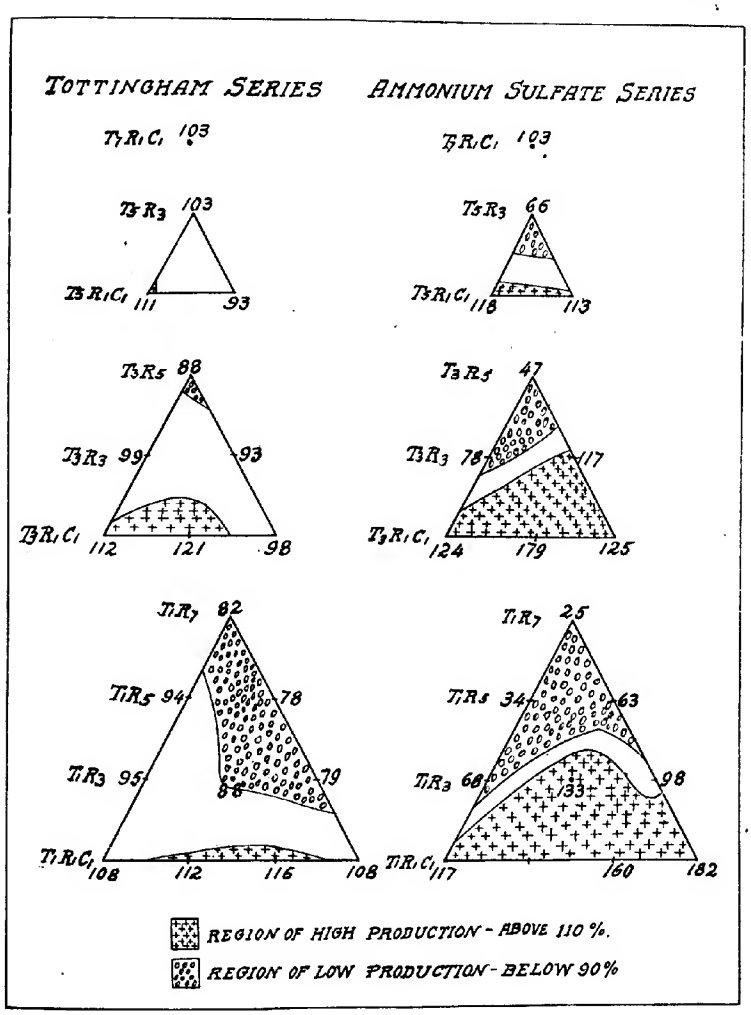

FIG. 5. THE RELATIVE AVERAGE VALUES FOR ROOT YIELDS OF SOYBEANS OF TOTTINGHAM AND AMMONIUM SULFATE SERIES

The high and low yield regions are shaded with crosses and circles, respectively.

as the poorest for the tops in the same series. The poorest culture in the Tottingham series was $T_1R_5C_3$, which was next to the poorest in the case of tops.

The region of high yields on the triangles for roots corresponds fairly well with the region of high yields on the triangles for tops; also, the areas of poor yields of roots approximately coincide with the areas of low yields of tops.

The ammonium sulfate series contains eleven cultures whose relative values are above 110, while only five cultures of the Tottingham series lie above that value. The relation is nearly reversed, however, when the number of cultures producing poor root yields is considered, or those whose relative values are below 90. In the ammonium sulfate series there are seven, but in the Tottingham series there are only five. It seems that the ammonium sulfate in the nutrient solution in the sand cultures caused a relatively better growth of

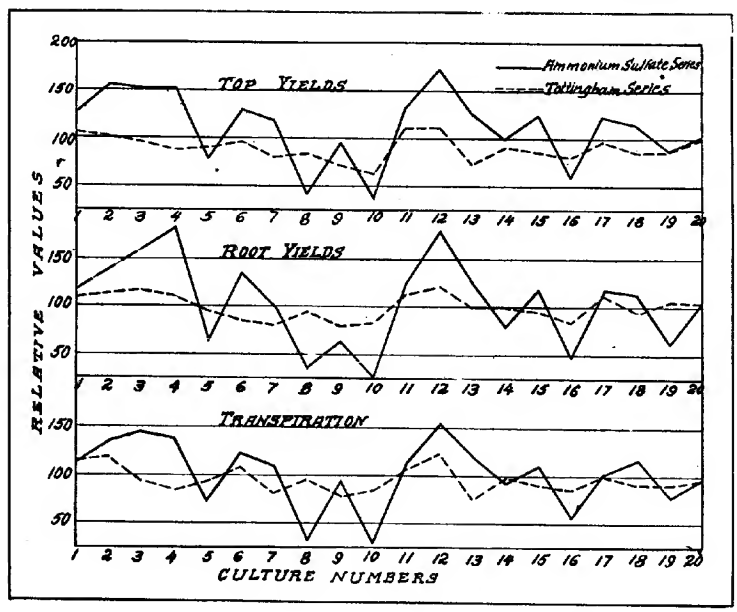

FIG. 6. RELATIVE VALUES FOR TOP YIELDS, ROOT YIELDS AND TRANSPIRATION OF WATER BY PLANTS OF SOYBEANS IN TOTTINGHAM AND AMMONIUM SULFATE SERIES

tops than of roots, while a reverse influence is noticed in the Tottingham series. This relation is plainly seen from the average figures of the relative values in tables 5 and 6, which are, in the ammonium sulfate series, 111 for tops and only 101 for roots. In the Tottingham series these figures are 89 and 99, respectively. In other words, the average root development in the different salt proportions of the ammonium sulfate series was quite large as compared with that of the Tottingham series. In this respect the data for roots and for tops show a very close agreement. Indeed, the relation between the growth of tops and that of roots in the same series is noticed throughout, as one can judge by studying the relative values in tables 5 and 6, and also in figure 6. With the

variation in the salt proportions from one culture to another, the curve for root yields of the ammonium sulfate series follows very closely the curve for the top yields of the same series. The relation between the root and top yields is also very similar in the Tottingham series, though these features are less pronounced than in the ammonium sulfate series. The correlation between the development of tops and that of roots is especially interesting, because it is not always found to exist with different plants. For instance, Tottingham (29), working with wheat in water cultures, could find no relation whatsoever between the dry weight of tops and that of roots. Shive (26) could trace some relations between yields of tops and roots. McCall (19) repeated Shive's work, carrying it along side of sand cultures, and published results that show very little correlation between the dry weight of tops and that of roots in both optimum series of Shive's water and sand cultures. Later, working with buckwheat in water cultures, Shive (27) found a fair relation between the growth of these two parts of the plant. Evidently, different plants behave differently as influenced by different salt proportions in the nutrient medium.

Transpiration and water requirement

The transpiration data were obtained by keeping a record of the water evaporated through the plants. In order to bring out clearly the relations between transpiration and the growth of tops and that of roots, the data of table 7 are presented. In both series the curves for transpiration data closely follow the curves of top yields and also of root yields. The relation of the transpiration of the roots to that of the tops, however, is more close in the case of the ammonium sulfate series than in that of the Tottingham series. In the case of the relation of transpiration to growth of tops, the results are well in accord with the results of some of the previous investigators, among whom could be mentioned Whitney and Cameron (31), Livingston, Britton and Reid (15), and especially Livingston (16 and 17), Shive (26) and McCall (19). Theoretically, the amount of water transpired, the moisture content of the soil and the meteorological conditions remaining the same, is mainly dependent upon both the leaf surface of the plant and the surface of the active root hairs. The influence of the leaf area, of course, is predominant. Some secondary factors, may play a prominent part only under some abnormal conditions of the plant growth.

The water requirement of the tops was calculated by dividing the total water loss in grams by the dry weight of tops. The data, together with their relative values, obtained by taking the average water requirement of both series as 100, are given in the table 7. The corresponding data for roots are presented in the same table. The results show the water requirement per gram of dry weight of tops in the Tottingham series to lie between 327 gm. in culture $T_3R_1C_1$ (the best culture for the top growth) and 387 gm. in culture

TABLE 7

Average transpiration, water requirement of tops and water requirement of roots, together with their relative values, of soybeans grown in Tottingham nutrient solution and the nutrient solution with ammonium sulfate in sand cultures

POSITION	NUMBER	TOTTINGHAM SERIES								NUMBER	AMMONIUM SULFATE SERIES							
		Transpiration				Tops		Roots			Transpiration				Tops		Roots	
		Trial 1	Trial 2	Average	Relative value	Grams of water required per gram of dry matter	Relative value	Grams of water required per gram of dry matter	Relative value		Trial 1	Trial 2	Average	Relative value	Grams of water required per gram of dry matter	Relative value	Grams of water required per gram of dry matter	Relative value
		grams	*grams*	*grams*							*grams*	*grams*	*grams*					
$T_1R_1C_1$	1	699	1156	926	114	369	108	2560	123	21	711	1110	911	112	298	87	2040	98
R_1C_3	2	780	1123	952	117	382	112	2240	108	22	737	1449	1093	134	287	84		
R_1C_5	3	735	798	767	94	344	101	1730	83	23	768	1477	1123	138	305	89	1845	89
R_1C_7	4	735	688	712	87	355	104	1730	83	24	748	1448	1093	134	292	85	2188	105
R_3C_1	5	570	964	767	94	360	105	2135	103	25	500	652	576	71	307	90	2223	107
R_3C_3	6	841	897	869	107	385	113	2595	125	26	709	1264	987	121	310	91	1945	94
R_3C_5	7	639	652	646	79	345	101	2143	103	27	665	1123	894	110	310	91	2420	117
R_5C_1	8	619	916	768	94	387	113	2160	104	28	438	92	265	32	285	83	2055	99
R_5C_3	9	687	573	630	77	380	111	2100	101	29	607	909	758	93	330	97	2950	142
R_7C_1	10	464	693	679	83	353	103	1853	89	30	366	136	251	31	192	56	2720	131
$T_3R_1C_1$	11	697	1028	863	106	327	96	1998	96	31	788	1062	925	114	294	86	1970	95
R_1C_3	12	823	1168	996	122	385	113	2170	104	32	921	1581	1251	154	298	87	1845	89
R_1C_5	13	611	563	587	72	342	100	1580	76	33	694	1233	964	118	316	92	2020	97
R_3C_1	14	673	869	771	95	366	107	2280	110	34	586	879	733	90	311	91	2515	121
R_3C_3	15	637	802	720	88	360	105	2085	100	35	723	1274	899	110	300	88	2025	98
R_5C_1	16	542	830	686	84	362	106	2065	99	36	416	475	446	55	328	96	2525	122
$T_5R_1C_1$	17	773	823	798	98	350	102	1895	91	37	771	870	821	101	378	81	1840	89
R_1C_3	18	758	712	735	90	370	108	2073	100	38	800	1095	948	116	354	104	2215	107
R_3C_1	19	592	866	729	90	356	104	1868	90	39	571	653	612	75	306	90	2450	118
$T_7R_1C_1$	20	678	853	766	94	324	95	1955	94	40	686	854	770	95	320	94	1975	95
Average						361	105.5	2060	99.3						322	94.5	2088	100.7

$T_1R_5C_1$. The average water requirement of the twenty duplicate cultures of this series for tops was 361 gm. In the ammonium sulfate series the corresponding values were 192 gm. for the lowest and 354 gm. for the highest, with cultures $T_1R_7C_1$ and $T_1R_1C_3$, respectively. Culture $T_1R_7C_1$ it should be remembered, gave the lowest yields of both tops and roots. The average water requirement of all the cultures of this series was 322 gm. The average value for both series was 342 gm., this value being taken as 100 in calculating the relative value for each culture.

The water requirement of the tops of the plants grown in the nutrient solution containing ammonium sulfate was considerably lower than the corresponding values for the plants grown in the solution of the Tottingham series. This is illustrated by figure 7, which also reveals the fact that the values of the water requirement for the two series run fairly parallel. Severely injured culture $T_1R_7C_1$ of the ammonium sulfate series is an exception to this general tendency. The curve for the Tottingham series was well above the other one.

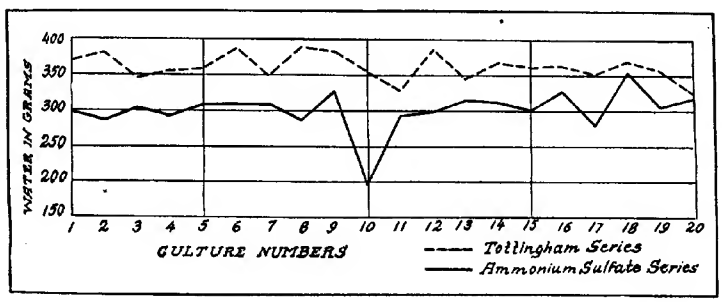

FIG. 7. WATER REQUIREMENTS IN GRAMS OF TOPS OF SOYBEANS PER GRAM OF DRY MATTER PRODUCED IN TOTTINGHAM AND AMMONIUM SULFATE SERIES

Again, as in the case of the yield of tops, the indication is clear that the nutrient solution containing ammonium sulfate, in comparison with the unmodified Tottingham solution, was better suited for the growth of soybeans.

In the experiments here recorded the total transpiration of water by the same number of plants in the ammonium sulfate series is sometimes more and sometimes less than in the case of Tottingham series, but it is always less per gram of dry weight of plants, so far as the yield of tops is concerned. No such consistent relation could be traced in the water requirement of the roots of the two series.

The water requirement of dry roots is, of course, many times as high as it is for tops. In the Tottingham series the values varied from 1580 gm. in culture $T_3R_1C_5$ to 2595 gm. in culture $T_1R_3C_3$, the average for all cultures being 2060 gm. The same values for the ammonium sulfate series were: lowest, 1840 gm. in culture $T_5R_1C_1$, and the highest, 2950 gm. in $T_1R_5C_3$, the average being 2.088 gm. The average water requirement of the roots for 40 duplicate

cultures was 2074 gm. Thus, the plants of the Tottingham series in the root development, as average figures show, were slightly more economical than the plants of the ammonium sulfate series.

COMPARISON OF YIELDS OF SOYBEANS GROWN IN NUTRIENT SOLUTIONS EMPLOYED BY OTHER AUTHORS

For the purpose of comparison, in the second trial of the experiment six additional cultures were prepared. Three duplicate cultures of the following solutions were employed: (a) Crone's, (b) Knop's and (c) Shive's. These

TABLE 8

Comparative growth of soybeans in sand cultures treated with nutrient solutions proposed by different authors

	Yield of tops		Yield of roots		Relative value of total yield	Transpiration		Water requirement of tops		Water requirement of roots	
	Dry weight	Relative value	Dry weight	Relative value		Amount	Relative value	Per gram of dry matter	Relative value	Per grams of dry matter	Relative value
	grams		*gram*			*grams*					
Crone 1	2.4092		0.5120			760					
Crone 2	2.4050		0.5108			758					
Average	2.4071	83	0.5114	100	183	759	72	371	95	1484	71
Knop 1	1.7975		0.3387								
Knop 2	1.6758		0.3862			825					
Average	1.7367	60	0.3625	71	131	825	79	475	121	2276	109
Shive 1	2.7605		0.4312			1088					
Shive 2	2.5455		0.4563			1069					
Average	2.6530	92	0.4438	87	179	1079	102	407	104	2431	116
Tottingham	2.7285	96	0.4510	88	184	1028	98	404	103	2279	109
Ammonium sulfate series	5.2335	182	0.7948	155	337	1581	150	302	77	1989	95
Average for all cultures	2.8791	100	0.5127	100	200	1054	100	392	100	2092	100

nutrient solutions had the following partial volume-molecular concentrations, each with a total concentration approximating an osmotic value of 2.5 atmospheres:

Crone's (4) solution: KNO_3, 0.0392 m.; $Ca_3(PO_4)_2$, 0.00314 m.; $Fe_2(PO_4)_2$, 0.0027 m.; $MgSO_4$, 0.008 m.; and $CaSO_4$, 0.007 m.

Knop's (13) solution: KNO_3, 0.0083 m.; KH_2PO_4, 0.0063 m.; $Ca(NO_3)_2$, 0.0207 m.; and $MgSO_4$, 0.0072 m.

Shive's (26) solution: KH_2PO_4, 0.0257 m.; $Ca(NO_3)_2$, 0.0074 m.; and $MgSO_4$, 0.0214 m.

The procedure of the experiment was exactly the same as in the main experiment. The results obtained with these nutrient solutions, together with

the data for the best yields from the Tottingham series and from the ammonium sulfate series, are presented in table 8.

An examination of the table 8 reveals the striking influence of the introduction of ammonium sulfate into Shive's nutrient solution, or of substituting it for KNO_3 in the Tottingham nutrient solution. The growth of soybeans in both the tops and the roots was very markedly increased.

Table 8 shows, in general, that transpiration was increased with the increase in growth. Also, the amount of water used per dry weight of crop was smaller in the case of plants of better development than in those of poor growth. On the whole, this is true of both the top and the root yields. Crone's nutrient solution deviated somewhat from this general rule.

DISCUSSION

In the foregoing experiments it was shown that ammonium sulfate, when added to Shive's nutrient solution, or when used as a substitute for potassium nitrate in Tottingham's nutrient solution, caused a greatly variable effect on the growth of both the tops and the roots of soybeans. In twenty representative cultures, of the 84 possible cultures used by Tottingham, ammonium sulfate in some salt proportions resulted in a very severe injury to the plants, while in other combinations it caused a remarkably high increase in the plant growth over the corresponding cultures of the Tottingham solution. Because of the complexity of the problem, it is often difficult (if not impossible) to determine why such profound differences occur in two cultures whose nutrient solutions differ but slightly. These questions were thoroughly discussed by Tottingham (29), Shive (26), and McCall (19) in the parallel cases and need not be repeated here. It is advisable, however, to trace the relations in connection with ammonium sulfate. Beginning with culture 21, or $T_1R_1C_1$, it will be observed (table 5) that with the smallest applications of mono-potassium phosphate, ammonium sulfate and calcium nitrate, when the partial osmotic concentrations supplied by magnesium sulfate was seven-tenths of the total concentration, the yield of the soybeans was well above the average. In the next culture, on the increase of calcium nitrate, the mono-potassium phosphate remaining the same, the yield increased about 13 per cent over the former. On the further increase of calcium nitrate and decrease in magnesium sulfate, the yield of tops did not increase, but was slightly decreased. A similar yield was obtained with the next culture having the highest application of calcium nitrate, although the roots (table 6) continued to increase. In culture 25, when the first increase in the amount of ammonium sulfate with the least amount of calcium nitrate was employed, the yield fell from 152 for culture 24 to 78 per cent for culture 25. The increase in calcium nitrate in the next culture to three-tenths of the total osmotic concentration brought the yield of plants to 132 per cent of the average. This concentration of calcium nitrate is the largest that could be safely used in combination with ammo-

nium sulfate in these experiments. In the next culture, No. 27, upon an increase in the calcium nitrate to five-tenths of the total osmotic concentration, a decrease in the yield of both tops and roots followed. The best yield of soybeans occurred in the culture with salt proportions as follows: 0.0156 m. mono-potassium phosphate, 0.0036 m. ammonium sulfate, 0.0108 m. calcium nitrate, and 0.0174 m. magnesium sulfate. On the reduction of calcium nitrate to one-tenth of the total osmotic concentration in culture 31, or on the increase of its proportion to five-tenths in culture 33, the decrease in the yield of tops was 30 per cent and 27 per cent, respectively. The difference effected in the case of yield of roots was still larger. It seems, therefore, that the development of the plant is the result of the combination, not of the different separate components of the nutrient solution. Tottingham (29), Shive (26), and later, McCall (19) have set forth the dependence of the separate salts upon the proportionality of the component factors in the solution, though the variation due to different salts in the instances of the above-mentioned investigators was not so pronounced as the variation due to difference in proportions of ammonium sulfate used in the present work.

It is commonly noticed that when an excess of ammonium sulfate is applied to an agricultural soil, an injury results to the plants grown therein. This injury varies according to the conditions under which the experiment is carried out. The amount of salt used, type of soil, amount of lime present (and also other fertilizers), moisture content, temperature, and kind of crop, are among the factors modifying the effect produced by ammonium sulfate in soil. In most cases, the ill effect of an excessive application of this salt is attributed to the acid production in the soil, when ammonium is used by the crop or converted into nitrate, leaving the acid radical behind. Another possibility, however, may be mentioned. It is a well established fact that there may be an injurious effect of a certain ion, as such, if its action is not antagonized by some other ion. McCool (20) reports that the NH_3-ion is very injurious above certain concentrations. Its ill effect, however, is diminished by the presence of other salts and can be completely destroyed by the antagonistic action of Ca or Na. In this connection it is interesting to note that the injurious effect of Na can be destroyed by the addition of a proper amount of NH_3. Though McCool dealt with the chlorides of these bases, the similar behavior of NH_3 in the form of sulfate can be expected. Although it is a common practice to correct the soil injured by excessive amounts of ammonium sulfate, with lime (CaO, $Ca(OH)_2$, $CaCO_3$, or $CaMg(CO_3)_2$), Schulze (28) in extensive field experiments counteracted the injurious effect of ammonium sulfate with the aid of sodium chloride, thus securing results similar to those which McCool obtained under better controlled laboratory conditions.

In certain salt proportions the ammonium sulfate is not only harmless to the development of the plants but extremely beneficial. Here lies the explanation of the fact that there are so many conflicting reports published regarding the effect produced by the action of this salt, when used as a fertilizer. It

seems that poorly balanced nutrient salt combinations account for the prejudice toward this salt as a source of nitrogen for plant growth.

That the differences caused by ammonium sulfate in the present experiments, observed from culture to culture, brought about by the change of salt proportions, were not due to the change in the concentration of the resultant solutions, is clearly brought out in table 3, which was previously discussed. Figure 3 also emphasizes the same point. The differences in the total osmotic concentration of the nutrient solution of the different salt proportions, in both the original form and after application to the sea sand, were not great enough to account for the large differences in the plant growth. Moreover, Ayres (1), working with tobacco in sand cultures, has shown that the total concentration, as measured by the total weight of salts in the initial application, is not as large a factor for the normal development of plants as the proper balance between the different salts applied. The preliminary work of McCall (19) also shows similar results.

CONCLUSION

In the experiments presented above, the work was done with soybeans grown in sand cultures.

The effect of ammonium sulfate on Shive's nutrient solution was studied. For this purpose ammonium sulfate was substituted for potassium nitrate in Tottingham's nutrient solution, which solution, as a control, was used in parallel with the ammonium sulfate series.

The osmotic concentration of the solutions was about 2.5 atmospheres, as calculated in advance.

The tested osmotic concentration of the sand in culture pots, using the freezing-point method, showed that the concentration of sand treated with the solution was nearly the same as that of the original solution.

With few exceptions all these values were below the calculated 2.5 atmospheres.

There was considerable difference between the action of Tottingham's nutrient solution and that containing ammonium sulfate in the place of potassium nitrate. In the case of the Tottingham nutrient solution the concentration of sand treated with solution in the laboratory was lower than the concentration of the original solution. On the other hand, in the sand treated with the nutrient solution containing ammonium sulfate, the concentration, with few exceptions, was higher than the concentration of the corresponding original solutions.

These phenomena were attributed to selective adsorption of ammonia, which, leaving the acid radical behind, resulted in the formation of acid, which later became more ionized than its salt, thus causing a greater depression of the freezing-point than could be expected from the salt solution.

The concentration values of the solutions, obtained by suction from the

pots at different periods of the experiment, did not vary considerably from those of the original solution. This indicated that the adsorption by sand was not the factor that could modify the concentration of the solution to such an extent as to cause even slight differences in plant growth.

Ammonium sulfate, when substituted for potassium nitrate, resulted in a better yield of soybeans in certain salt proportions, than in the corresponding salt proportions with potassium nitrate, but caused considerable injury when added in excess.

The foliage of the plants receiving ammonium sulfate, on the whole, had a greener color than that of the plants in the Tottingham series.

The best cultures for the growth of tops in the Tottingham series were $T_3R_1C_1$ and $T_3R_1C_3$, corresponding to 0.0156 m. mono-potassium phosphate, 0.0049 m. potassium nitrate, 0.0036 m. calcium nitrate, and 0.0290 m. magnesium sulfate in the former, and 0.0156 m. mono-potassium phosphate, 0.0049 m. potassium nitrate, 0.0108 m. calcium nitrate and 0.0174 m. magnesium sulfate in the latter.

The best cultures of the ammonium sulfate series was $T_3R_1C_3$, which contained 0.0156 m. mono-potassium phosphate, 0.0036 m. ammonium sulfate, 0.0188 m. calcium nitrate and 0.0174 m. magnesium sulfate.

The best culture of the ammonium sulfate series gave a dry weight of tops 35 per cent higher than that of the best culture in the Tottingham series.

In the case of top yields, only two cultures in the Tottingham series gave results 110 per cent or more of the average, while twelve cultures gave 90 per cent or less than 90 per cent of the average. In the ammonium sulfate series, twelve cultures were above and five cultures below this average.

The variations from culture to culture with the change in the salt proportions were much greater in the ammonium sulfate series than in the Tottingham series.

Similar relations exist, in general, in the case of root development. However, more cultures in the ammonium sulfate series gave root yields below 90 per cent of the average than the number of cultures in the same series that gave top yields below 90 per cent of the average.

There is a very close relation between the yield of tops, yield of roots, and the total transpiration of the plants. Increase in the yield of tops was followed by an increase in the yield of roots, and both these values were accompanied by increased transpiration.

The water requirement (water required to produce 1 gm. of dry matter) of soybean tops in the cultures of the ammonium sulfate series was less than that in the corresponding cultures of the Tottingham series.

In comparing the growth of soybeans in sand cultures treated with different solutions, as proposed by other authors, the order of magnitude of yields of tops was as follows: ammonium sulfate series > Tottingham > Shive R_5C_2 > Crone > Knop.

The author wishes to acknowledge his indebtedness to Dr. John W. Shive

(in whose laboratory the work has been done) for suggesting the problem, and for helpful advice, ever ready at the author's disposal during the progress of the work. The author is also indebted to Dr. J. G. Lipman for the interest he has taken in the work, and for his assistance in interpreting the data.

REFERENCES

(1) AYRES, H. A. 1917 Influence of the composition and concentration of the nutrient solution on plants grown in sand cultures. Univ. Cal. Pub. Agr. Sci., v. 1, no. 11, p. 341–394.

(2) BOUYOUCOS, G. J., AND MCCOOL, M. M. 1915 The freezing point method as a new means of measuring the concentration of the soil solution directly in the soil. Mich. Agr. Exp. Sta. Tech. Bul. 24.

(3) BREAZEALE, J. E. 1905 Effect of the concentration of the nutrient solution upon wheat cultures. *In* Science, v. 22, p. 146–149.

(4) CRONE, G. 1902 Ergebnisse von Untersuchungen über die Wirkung der Phosphorsaure auf die höhere Phlanzen und eine neue Nahrlosung. Sitzber. Gesell. Natur u. Heilk. Bonn, (1902), p. 167–173.

(5) FREAR, WILLIAM 1915 Sour soils and liming. Penn. Dept. Agr. Bul. 261.

(6) GERLACH AND VOGEL 1905 Ammoniacal nitrogen as a plant food. *In* Centbl. Bakt. (etc.), Abt. 2, Bd. 14, No. 3, 4, p. 124–128.

(7) GORE, G. 1894 Decomposition of liquids by contact with powdered silica. *In* Chem. News, v. 69, p. 22–24, 33, 44–46.

(8) GRAHAM, THOMAS 1830 Von den Wirkungen der thierischen Kohle auf verschiedene Losingen. *In* Poggendorff Ann. Phys. u. Chem., Bd. 19, p. 139–145.

(9) HARRIS, F. S. 1914 Effect of variation in moisture content on certain properties of soil and on the growth of wheat. N. Y. (Cornell) Agr. Exp. Sta. Bul. 352.

(10) HARRIS, J. E. AND GORTNER, R. A. 1914 Notes on the calculation of the osmotic pressure of expressed vegetable saps from the depression of the freezing point, with a table for the values of P from = 0.001° to = 2.999°. *In* Amer. Jour. Bot., v. 1, p. 75–78.

(11) HELLRIEGEL, H. 1883 Beitrage zu den naturewissenschaftlichen Grundlagen des Ackerbaues Braunschweig. Cited by Russel, E. J., in Soil Conditions and Plant Growth, p. 27, 1915.

(12) JONES, H. C. 1912 The electrical conductivity, dissociation and temperature coefficients of conductivity from zero to sixty-five degrees, of aqueous solutions of a number of salts and organic acids. Carnegie Inst. Washington Pub. 170.

(13) KNOP, W. 1862 Quantitative-analytische Arbeiten über den Ernährungsprocess der Pflanzen. II. *In* Landw. Vers. Stat., Bd. 4, p. 173–187.

(14) LAGERGREN, S. 1898 Zur Theorie der Sogenannten Adsorption Gelöster Stoffe. *In* Bihangtill K. Svenska Vet. Akad. Handlinger, v. 24, no. 4, p. 3–39.

(15) LIVINGSTON, B. E., BRITTON, J. C., AND REID, F. R. 1905 Studies on the properties of an unproductive soil. U. S. Dept. Agr. Bur. Soils, Bul. 28.

(16) LIVINGSTON, B. E. 1905 Relation of transpiration to growth of wheat. *In* Bot. Gaz. v. 40, p. 178–195.

(17) LIVINGSTON, B. E. 1907 Further studies on the properties of unproductive soils; U. S. Dept. Agr. Bur. Soils Bul. 36.

(18) MCCALL, A. G. 1916 Adsorption of potassium by the soil. *In* Jour. Phys. Chem., v. 20, no. 1, p. 51–63.

(19) MCCALL, A. G. 1916 Physiological balance of nutrient solutions for plants in sand cultures. *In* Soil Sci., v. 2, no. 3, p. 207–254.

(20) McCool, M. M. 1913 The action of certain nutrient and non-nutrient bases on plant growth. N. Y. (Cornell) Agr. Exp. Sta. Memoir 2.

(21) Nikolaeva, A. G. 1914 The formation of asparagin by Lupinus luteus. *In* Iz Rezult. Veget. Opytov Lab. Rabot, Moscov. Selsk. Khoz. Inst. (Ann. Inst. Agron. Moscow), v. 10, sup. 3, p. 380–383.

(22) Osterhaut, W. J. V. 1909 The nature of balanced solutions. *In* Bot. Gaz., v. 47, p. 148.

(23) Prianishnikov, D. N. 1900 The influence of the moisture of the soil on the growth of plants. *In* Zhur. Opuitn. Agron. [Russ. Jour. Expt. Landw.], no. 1, p. 3–20.

(24) Prianishnikov, D. N. 1914 The role of ammonia in the transformation of nitrogenous material in plants. *In* Iz Rezult. Veget. Opytov Lab. Rabot, Muscov. Selsk. Khoz. Inst. (Ann. Inst. Agron. Moscow), v. 10, sup. 3, p. 24.

(25) Sachs, J. 1860 Vegetationsversuche mit Ausschluss des Bodens über die Nahrstoffe und Sonstigen Ernährungsbedingungen von Mais Bohnen und anderen Pflanzen. *In* Landw. Vers. Sta., Bd. 2, p. 219–268.

(26) Shive, J. W. 1915 A study of physiological balance in nutrient media. *In* Physiol. Researches, v. 1, no. 7, p. 327–397.

(27) Shive, J. W. 1917 A study of physiological balance for buckwheat grown in three-salt solutions. N. J. Agr. Exp. Sta. Bul. 319.

(28) Schulze, B. 1910 The fertilizer action of salt in combination with ammonium sulfate. *In* Mitt. Deut. Landw. Gesell, Bd. 25, No. 30, p. 452–458.

(29) Tottingham, W. E. 1914 A quantitative chemical and physiological study of nutrient solutions for plant cultures. *In* Physiol. Researches, v. 1, no. 4, p. 133–245.

(30) Tulaikoff, N. 1915 Transpiration coefficient of cultural plants. *In* Zhur. Opuitn. Agron. [Russ. Jour. Expt. Landw.], v. 16, no. 1, p. 36.

(31) Whitney, M., and Cameron, F. K. 1904 Investigations in soil fertility. U. S. Dept. Agr. Bur. Soil. Bul. 23.

(32) William, A. M. 1914 On "negative" adsorption. *In* Trans. Faraday Soc., v. 10, part 1, p. 155–159.

(33) Wolkoff, M. I. 1917 Adsorption of ammonium sulfate of different concentrations by different salts and quartz sand of different fineness. *In* Soil Sci., v. 3, no. 6, p. 561–564.

CARBONIC ACID GAS IN RELATION TO SOIL ACIDITY CHANGES

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During the past two years four articles have been published which deal with certain phases of recent work on soil acidity. The articles are:—"Acid Soils and the Effect of Acid Phosphate upon Them" by S. D. Conner; (2) "Acidity and Absorption as Measured by the Hydrogen Electrode," by Sharp and Hoagland (6); "The Reaction of Soil and Measurements of Hydrogen Ion Concentration" by Gillespie (5) and "Liming and Lime Requirement of Soil," by Ames and Scholenberger (1). Among the recent workers on and contributors to theories of soil acidity may be mentioned Cornu, Daikuhara, Frear, Gans, Harris, MacIntire, Morse, Parker, Rice, Ruprecht, Saidel, Sullivan, Truog and Veitch.

Acidity of soils is corrected by additions of lime. One of the most generally recommended forms of lime to apply is finely-ground calcium limestone containing as high as 44 per cent of carbon dioxide. All good agricultural practices involve the addition of organic matter to the soil. Soil organic matter is decomposed by bacteria with the production of immense amounts of carbon dioxide, and thus the effect of carbon dioxide upon soil acidity is worthy of investigation. The work of Coville (3, 4) and others has shown that organic matter is acid in reaction at certain stages of its decay. We are aware of no work that absolutely proves that this acidity is other than that due to carbonic acid weakly held by the organic matter.

The experiments reported upon here were designed primarily to obtain data on the effect of carbon dioxide additions to soil in relation to soil and plant

*Reaction of minerals after extraction by carbon dioxide solution**

ROCK OR MINERAL	ORIGINAL REACTION	REACTION AFTER TREATMENT WITH CO_2	
		With water	After addition of KCl solution
Granite	Alkaline	Weakly acid	Weakly acid
Gneiss	Alkaline	Weakly acid	Acid
Hornblende Andesite	Weakly alkaline	Weakly acid	Acid
Basalt	Strongly alkaline	Weakly acid	Weakly acid
Feldspar	Strongly alkaline	Weakly acid	Weakly acid
Mica	Strongly alkaline	Acid	Acid

* From Ames, J. W. and Schollenberger, C. J. (1, p. 331).

changes. The acidity data obtained were reported in a separate paper since they give additional information confirmatory of the work of Daikuhara which has been quoted in many articles on soil acidity.

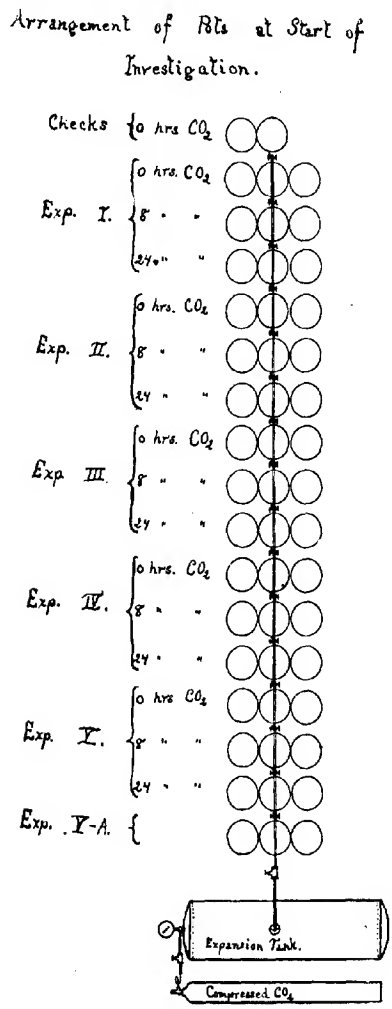

FIG. 1. SYSTEM OF TREATMENT

PLAN OF THE INVESTIGATION

Equal weights of soil were put into paraffined Wagner pots of the most approved type, after the soil had been thoroughly mixed and specified applications of fertilizer and lime added. The soil was compacted uniformly in

the pots by dropping them a prescribed number of times. Distilled water was added to bring the moisture content up to half saturation and small pepper plants were transplanted into the pots. The soil was kept with a dust mulch and uniform moisture content throughout the entire investigation, extending from February 4 to December 5, 1916. The position of the pots was changed from time to time in such a way that each pot was in a position occupied by every other pot at least twice during the period of investigation.

In each experiment there were nine pots—three which received no carbon dioxide application, three where the gas bubbled into the soil between 8.00 a.m. and 4.00 p.m. and three where the gas was bubbled constantly into the soil. Figure 1 shows the arrangement of the different experiments on the

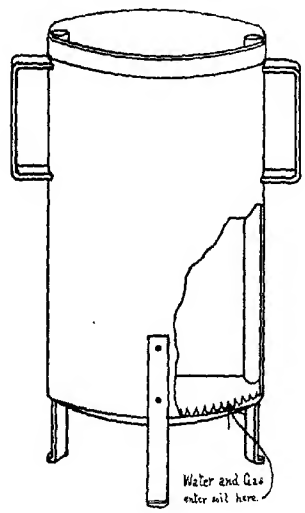

FIG. 2. WAGNER POT

greenhouse bench, figure 2 shows a Wagner pot. A set of pots is shown in plate 1. Carbon dioxide applications were made to the soil from April 16 to the close of the investigation. The gas was applied, to each pot at the rate of approximately 650 cc. (under standard conditions) of gas per hour of treatment given.

ACIDITY METHODS USED

In making acidity tests two methods were used—that recommended by Veitch and that recommended by Hopkins, Pettit and Knox. In one experiment lime was added to satisfy the lime requirement as determined by the Veitch method; in another case one-half this amount was used; and in a third instance one and one-half times the lime requirement was applied. In both

methods special care was taken in titrating. The potassium nitrate used for the Hopkins method was neutral to phenolphthalein.

Analysis of soil

	Per cent
Insoluble residue (1)	82.05
K_2O (1)	0.41
Na_2O (1)	0.36
CaO (1)	0.44
MgO (1)	0.79
Fe_2O_3 (2)	4.35
Al_2O_3 (1)	3.65
P_2O_5 (3)	0.13
SO_2 (1)	0.52
Water (4)	2.31
Volatile matter (4)	5.76
Nitrogen (5)	0.19
Total carbon (6)	2.12
Inorganic carbon (7)	0.03

(1) Solution and residue made by extracting soil on steam bath with hydrochloric acid (specific gravity 1.115) for 10 hours. Solution analyzed by usual procedures.

(2) Determined volumetrically from acid solution with hydrogen sulfide as reducing agent.

(3) Method of Goss.

(4) Five-gram aliquot used and both determinations made on same aliquots.

(5) Regular Kjeldahl.

(6) Method of Ames and Gaither as modified by Schollenberger.

(7) Method of Marr with dilute acid and vacuum as recommended by Ames of Ohio.

Table 1 and figure 3 give the soil acidity as influenced by cropping, fertilization, lime, and carbon dioxide. The figures express parts of calcium carbonate per hundred parts of dry soil as determined by the Hopkins potassium nitrate method. It is noted that the acid soil increases in acidity when allowed to stand in the greenhouse with its water-holding capacity one-half satisfied. The increase in acidity of 350 pounds per million pounds of soil was more than the acidity of the soil at the start of the investigation. When the soil was cropped in experiment 1 an increase in acidity, slightly greater than where there was no crop, resulted. The carbon dioxide applications increased the acidity further. The constant treatment of carbon dioxide gave the greater increase in acidity.

In experiment 2 we note that the lime without carbon dioxide gas prevented as large an increase in acidity, whereas carbon dioxide gas applications with lime gave greater increases in acidity.

In experiment 3 the soil was fully neutralized according to the Veitch method, but gave almost as great an increase in acidity as where one application of lime was used. The extra application of lime prevented the increases in acidity due to carbon dioxide treatments from being as great.

In experiment 4 the triple application of lime and one application of phosphorus gave smaller acidity increases where no carbon dioxide was applied.

TABLE 1

*Soil acidity as influenced by cropping, fertilization, liming and carbon dioxide**

	CO_2 TREATMENT GIVEN PER DAY	$CaCO_3$ ADDED	ACIDITY AT END OF INVESTIGATION (HOPKINS METHOD)	TOTAL INCREASE IN ACIDITY (HOPKINS METHOD)	INCREASE DUE TO CO_2
	hours				
Acid soil, no crop	0	0.0000*	0.0583*	0.0350*	
Experiment 1					
Acid soil, with crop	0	0.0000	0.0592	0.0359	
	8	0.0000	0.0716	0.0483	0.0124†
	24	0.0000	0.0881	0.0648	0.0289
Experiment 2					
Acid soil, with crop, with single application of $CaCO_3$	0	0.0770	0.0362	0.0129	
	8	0.0770	0.0644	0.0411	0.0282
	24	0.0770	0.0736	0.0503	0.0374
Experiment 3					
Acid soil, with crop, with double application of $CaCO_3$	0	0.1540	0.0353	0.0120	
	8	0.1540	0.0449	0.0216	0.0092
	24	0.1540	0.0510	0.0277	0.0157
Experiment 4†					
Acid soil, with crop, with triple application of $CaCO_3$ and single application of phosphorus Banner bone (di-calcium phosphate)	0	0.2310	0.0242	0.0009	
	8	0.2310	0.0350	0.0117	0.0108
	24	0.2310	0.0333	0.0100	0.0091
Acid phosphate	0	0.2310	0.0324	0.0091	
	8	0.2310	0.0456	0.0223	0.0132
	24	0.2310	0.0416	0.0183	0.0092
Experiment 5‡					
Acid soil, with crop, with triple application of $CaCO_3$ and single application of nitrogen Dried blood	0	0.2310	0.0315	0.0082	
	8	0.2310	0.0397	0.0164	0.0082
	24	0.2310	0.0454	0.0221	0.0139
Sodium nitrate	0	0.2310	0.0321	0.0088	
	8	0.2310	0.0321	0.0088	0.0000
	24	0.2310	0.0475	0.0242	0.0154

TABLE 1—(Continued)

Experiment 5A §

	CO_2 TREATMENT GIVEN PER DAY	$CaCO_3$ ADDED	ACIDITY AT END OF INVESTIGATION (HOPKINS METHOD)	TOTAL INCREASE IN ACIDITY (HOPKINS METHOD)	INCREASE DUE TO CO_2
Acid soil, with crop, with triple application of $CaCO_3$ and approximately one-half application of nitrogen in form of sodium nitrate	0	0.2310	0.0227	0.0004	
	8	0.2310	0.0318	0.0085	0.0081
	24	0.2310	0.0365	0.0132	0.0128

* Single application of lime equals 770 parts $CaCO_3$ per million of dry soil.

Single application of phosphorus equals 501.5 parts per million of dry soil.

Single application of nitrogen equals 235.6 parts per million of dry soil.

Acidity of soil used for investigation 0.0233 (by Hopkins) and 0.1540 (by Veitch) parts of $CaCO_3$ per 100 parts of dry soil.

† Figures express parts $CaCO_3$ per 100 parts of dry soil.

‡ In experiments 4 and 5 comparisons were made between different carriers of phosphorus and nitrogen.

Two pots in each set of three received Banner Bone and one acid phosphate in experiment 4.

Two parts in each set of three received dried blood and one sodium nitrate in experiment 5.

§ CO_2 treatment started 20 weeks later than in other experiments.

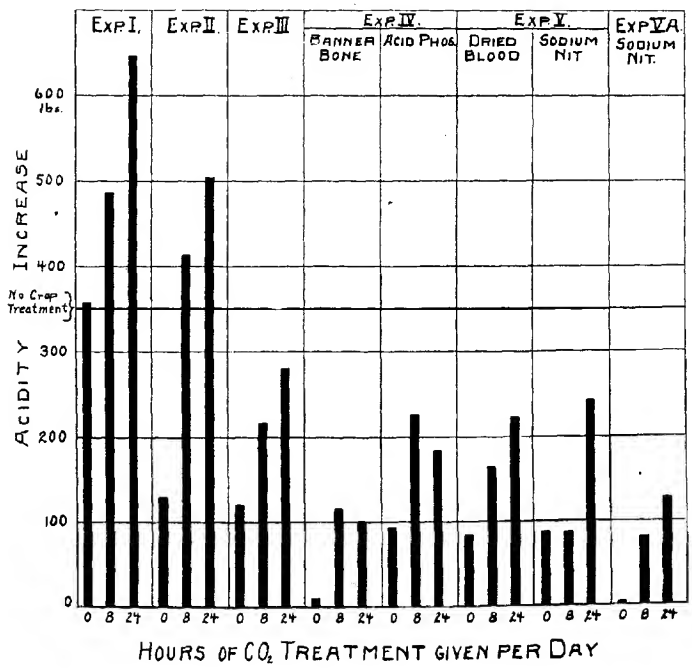

FIG. 3. ACIDITY CHANGES DUE TO CROPPING, LIMING, FERTILIZATION AND CARBON DIOXIDE TREATMENTS

This might have been due to the extra application of calcium carbonate. The increases in acidity with Banner Bone were much less than where acid phosphate was used. The increases in acidity due to carbon dioxide applications were less with both phosphorus carriers under constant carbon dioxide treatments than they were with the intermittent carbon dioxide treatments. The increases due to carbon dioxide were practically the same with Banner Bone as they were with acid phosphate.

The effects of carbon dioxide in increasing soil acidity where dried blood and sodium nitrate were compared as nitrogen carriers are given under experiment 5. With no carbon dioxide treatment the increases in soil acidity were practically the same for dried blood and sodium nitrate. With the intermittent carbon dioxide treatment there was practically double the total increase with dried blood that there was with sodium nitrate. With the constant carbon dioxide treatment the total increase was slightly greater where sodium nitrate was used. This made the increases due to carbon dioxide practically the same where constant carbon dioxide treatments were given, but no increase and 82 pounds increase with the intermittent gas treatments.

Experiment 5-A is a test of one pot in each case to determine if results would be greatly different if smaller amounts of sodium nitrate were used. It is noted that there is practically no increase in acidity without the carbon dioxide treatments but that acidity is increased by both carbon dioxide applications.

Figure 4 gives the acidity titrations in cubic centimeters of N/10 sodium hydroxide in relation to the weights of ignited precipitates from the acidity solutions. These precipitates were obtained from the hot solutions by adding ammonium hydroxide after the sodium hydroxide precipitates had been dissolved with hydrochloric acid and the solutions boiled. This graph would be a continuous curve or straight line instead of an oscillating graph if the treatments given this soil changed its acidity in proportion as the potassium-nitrate-soluble iron and aluminum changed. The graph shows that the Hopkins potassium nitrate method gives acidity results that can not be correlated with the amount of aluminum in the soil that is in solution in presence of a normal salt of a strong acid and strong base.

The ignited precipitates were composited by carbon dioxide treatments so that the effect of carbon dioxide applications on the composition of the precipitate could be determined. Weights and analyses of precipitates are given in table 2.

There are many theories as to the nature of soil acidity, which may be classified in two groups: physical and chemical. Among the physical theories we have one depending upon what has been termed "selective adsorption," where the soil is supposed to adsorb the basic portion of the compound. Of the chemical theories offered we find an explanation based on the existence of free acids in soils as the resultant of hydrogen-ion determinations and mass-action studies. In fact, some chemical theories of soil acidity take into account the relative velocity of different chemical reactions. The data given

TABLE 2

Weights and analyses of ignited precipitates

	CARBON DIOXIDE TREATMENT PER DAY		
	0 hours	8 hours	24 hours
Total weight of precipitate analyzed (gm.)	0.2129*	0.2331	0.2362
Silicon dioxide—SiO_2 (per cent)	9.34	7.51	8.68
Ferric oxide—Fe_2O_3 (per cent)	1.32	1.09	1.28
Aluminum oxide—Al_2O_3 (by difference) (per cent)	89.34	91.40	90.04

* Includes precipitate from pot receiving no crop and no CO_2 treatments.

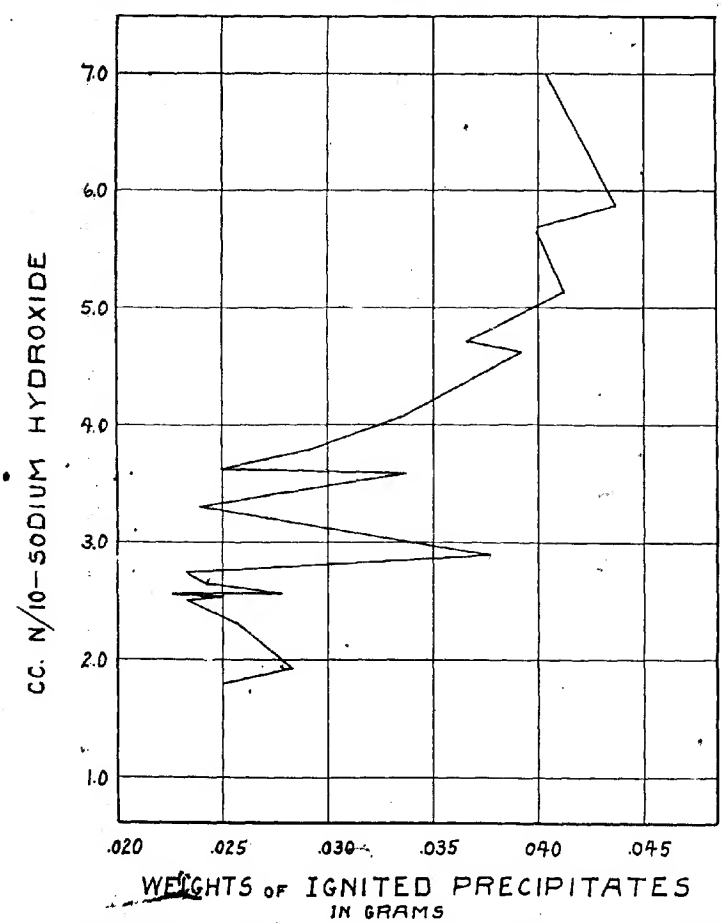

FIG. 4. RELATION BETWEEN ACIDITY TITRATIONS AND WEIGHT OF IGNITED MATERIAL PRECIPITATED FROM ACIDITY SOLUTIONS BY AMMONIUM HYDROXIDE

in this paper support the chemical theories of soil acidity, inasmuch as different applications of a gas of definite composition (carbon dioxide), which is not only soluble in water but which combines with it, yielding hydrogen ions, caused differences in soil acidity. These differences in soil acidity have varied with different fertilizer and lime applications, both with and without carbon dioxide treatments. This leads us to conclude that the figures given by the Hopkins potassium nitrate method show that liming, fertilization, and carbon dioxide applications, both alone and in combination, have brought about different chemical reactions in the soil used.

CONCLUSIONS

1. Keeping soil at one-half its water-holding capacity in a greenhouse increased its acidity.
2. Cropping soil kept at one-half its water-holding capacity increased its acidity.
3. The increases in acidity of cropped soil were modified by different applications of calcium carbonate.
4. The increases in soil acidity of cropped soil varied with different fertilizer applications.
5. Carbon dioxide added to cropped soil, treated with lime alone or lime and fertilizer, increased its acidity.
6. The results of these experiments support chemical theories as to the nature and causes of soil acidity.
7. The changed reactions of this soil towards a neutral salt of a strong acid and a strong acid (KNO_3) after subjection to the varied conditions of the experiment at least suggests that soil acidity is largely the result of hydrolytic mass action phenomena.

The authors wish to make acknowledgment to Director C. G. Woodbury for permission to carry on this work.

REFERENCES

(1) Ames J. W., and Schollenberger, C. J. 1916 Liming and lime-requirement of soil. Ohio Agr. Exp. Sta. Bul. 306.

(2) Conner, S. D. 1916 Acid soils and the effect of acid phosphate and other fertilizers upon them. *In* Jour. Indus. Engin. Chem., v. 8, no. 1, p. 35–40.

(3) Coville, F. V. 1910 Experiments in blueberry culture, U. S. Dept. Agr. Bur. Plant Indus. Bul. 193.

(4) Coville, F. V. 1914 The formation of leaf mould. *In* Smithsonian Report, 1913, p. 333–343.

(5) Gillespie, L. J. 1916 The reaction of soil and the measurements of hydrogen-ion concentration. *In* Jour. Wash. Acad. Sci., v. 6, no. 1, p. 7–16.

(6) Sharp, L. T., and Hoagland, D. R. 1916 Acidity and adsorption in soils as measured by the hydrogen electrode. *In* Jour. Agr. Research, v. 7, no. 3, p. 123–145.

PLATE I

Pots Connected with Carbon Dioxide Line for Carbon Dioxide Applications to Soil

THE NITROGEN DISTRIBUTION OF FIBRIN HYDROLYZED IN THE PRESENCE OF FERRIC CHLORIDE

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INTRODUCTION

In a recent investigation of the distribution of the organic nitrogen in the soil Morrow and Gortner (5) made the statement that "the humin nitrogen of protein origin actually present in the hydrolyzed soil may easily be a very small part of the nitrogen found." It was shown that from 3.26 to 9.21 per cent of the total nitrogen was precipitated by calcium hydroxide. This did not represent *true* humin nitrogen since the calcium hydroxide precipitate did not contain any black substance formed by hydrolysis. The solution from which it was precipitated was colored only with ferric compounds. They therefore concluded that the organic material in this precipitate must consist of colorless organic compounds absorbed by or combined with the lime and that the nitrogen contained in this fraction must consist almost entirely of material of non-protein origin since in pure proteins the nitrogen retained in the calcium hydroxide precipitate is supposed to consist entirely of deeply colored substances.

Gortner (2) has shown that when fibrin is hydrolyzed in the presence of varying amounts of carbohydrates the humin nitrogen shows an increase. This indicates that a pure protein when hydrolyzed in the presence of an oxidizing agent such as a carbohydrate causes a redistribution of the fractions with an increase in the humin nitrogen.

EXPERIMENTAL

The problem

We have made a study of the distribution of the nitrogen of a pure protein hydrolyzed in the presence of ferric chloride as an oxidizing agent. This compound was chosen because all mineral soils contain compounds of iron and it was thought that additional data might be obtained which would throw more light on the formation of humin nitrogen.

The material

The study has been made on a pure protein. The protein selected was Merck's fibrin from blood.

The method

Duplicate analyses were made with fibrin and fibrin plus ferric chloride. In each case 3 gm. of fibrin were hydrolyzed in the presence of HCl (sp. gr. 1.115) for 48 hours at gentle boiling. The calculated amount of hydrated ferric chloride equivalent to 30 gm. of anhydrous ferric chloride was added to one set of duplicates. The calculated amount of concentrated hydrochloric acid was also added to the flasks containing the ferric chloride, so that the

TABLE 1

Comparative analyses of 3 gm. of fibrin hydrolyzed alone and in the presence of 30 gm. of anhydrous ferric chloride

NITROGEN	3 GM. FIBRIN, NO $FeCl_3$					3 GM. FIBRIN + 30 GM. $FeCl_3$				
	Nitrogen (mgm.)		Per cent of total N			Nitrogen (mgm.)		Per cent of total N		
	I	II	I	II	Average	I	II	I	II	Average
Total	0.4538	0.4444				0.4546	0.4617			
Ammonia	0.0473	0.0468	10.42	10.53	10.48	0.0589	0.0618	12.95	13.39	13.17
Acid-insoluble humin	0.0095	0.0096	2.09	2.16	2.13	0.0075	0.0077	1.65	1.67	1.66
Acid-soluble humin precipitated by $Ca(OH)_2$	0.0047	0.0043	1.04	0.97	1.00	0.0459	0.0443	10.09	9.59	9.84
Phosphotungstic acid humin	0.0010	0.0046	0.22	1.04	0.64	0.0046	0.0036	1.01	0.78	0.90
Total humin	0.0152	0.0185	3.35	4.16	3.76	0.0580	0.0556	12.76	12.04	12.40
Basic	0.1143	0.1230	25.19	27.68	26.44	0.1114	0.1072	24.50	23.21	23.86
Arginine	0.0627	0.0598	13.81	13.46	13.63	0.0615	0.0605	13.53	13.10	13.31
Histidine	None	0.0023	None	0.52	0.26	None	None			None
Lysine	0.0499	0.0585	11.00	13.16	12.08	0.0464	0.0434	10.21	9.40	9.81
Cystine	0.0017	0.0024	0.37	0.54	0.46	0.0035	0.0033	0.77	0.71	0.74
Amino, in bases	0.0700	0.0673	15.43	15.15	15.29	0.0688	0.0709	15.13	15.36	15.25
Non-amino, in bases	0.0443	0.0557	9.76	12.53	11.15	0.0426	0.0363	9.37	7.86	8.61
Filtrate from bases	0.2726	0.2671	60.06	60.10	60.08	0.2346	0.2333	51.59	50.54	51.07
Amino, in filtrate from bases	0.2500	0.2512	55.08	56.52	55.80	0.2077	0.2123	45.68	45.97	45.82
Non-amino, in filtrate from bases	0.0226	0.0159	4.98	3.58	4.28	0.0269	0.0210	5.92	4.55	5.24
Total recovered	0.4494	0.4554	99.02	102.48	100.75	0.4629	0.4579	101.85	99.17	100.51

water of crystallization in the salt would not reduce the acid below constant boiling. The resulting hydrolysates were analyzed according to Van Slyke's (6, 7) method. Instead of the single humin nitrogen fraction we have found it desirable to follow the suggestion of Gortner and Holm (3) and separate the humin nitrogen into three fractions (a) acid-insoluble humin; (b) acid-soluble humin [precipitated by $Ca(OH)_2$] and (c) phosphotungstic acid humin (precipitated by phosphotungstic acid).

The "acid-insoluble humin" was obtained by first diluting the cold hydrolystate with about an equal volume of water, filtering off the insoluble humin and washing until free of chlorides. A Kjeldahl determination gave the nitrogen content. After the removal of the acid-insoluble humin the acid filtrate was evaporated under diminished pressure until all the hydrochloric acid possible was driven off. Water, alcohol and a suspension of calcium hydroxide were added and the ammonia determined in the usual manner. In the determination of the ammonia nitrogen from the fibrin plus ferric chloride, however, the distillation was continued for an hour, since it has been shown by one of us (5) that all the ammonia nitrogen was not driven off in a half-hour when the volume of the solution was large and a bulky precipitate of hydroxide was present. The precipitate remaining in the distilling flask was filtered and washed free of chlorides. The samples to which ferric chloride was added were washed by decantation after the method previously described (5) for mineral soils. A Kjeldahl determination was made and the results recorded as "acid-soluble humin." The filtrate from the acid-soluble humin was concentrated and diluted to 250 cc. volume. Two 25-cc. portions were used for the determination of the total nitrogen in the solution. The remaining 200 cc. of the solution were used for the precipitation of the diamino acids. The precipitation, washing and the decomposition were carried out according to Van Slyke's directions. The washed precipitate of barium phosphotungstate was subjected to Kjeldahl analysis and the nitrogen was recorded as "phosphotungstic acid humin." The remainder of the analysis was completed as directed by Van Slyke. All titrations were made with N/14 acid and alkali so that the figures obtained represented milligrams of nitrogen without the necessity of a calculation.

Analytical data

The distribution of the nitrogen both as regards grams of nitrogen in the different fractions and the percentages of the total nitrogen is shown in table 1.

DISCUSSION

It is observed from a study of the table that the hydrolysis carried out in the presence of ferric chloride gives a larger proportion of the nitrogen in the fraction precipitated by calcium hydroxide than with the fibrin hydrolyzed alone. This increase in the acid-soluble humin is of the same order as that previously found for the hydrolysate of soils and in this instance corresponds very closely to the decrease in the filtrate from the bases.

In all probability the humin nitrogen precipitated by the calcium hydroxide from the soil hydrolysates was largely protein nitrogen instead of being of non-protein origin. There is no doubt, however, that some of this fraction must be due to non-protein material. Gortner (1) has shown that uric acid

nitrogen is distributed in all four of the major fractions after hydrolysis. Purine and pyrimidine bases are undoubtedly present in the soil organisms and a certain part of their nitrogen would be precipitated in the fraction under discussion.

It seems evident from this investigation that the earlier conclusions of one of us (5) in regard to the humin nitrogen precipitated by calcium hydroxide from the soil hydrolysate were in part in error, and that in all probability a part of these colorless substances are derived from the filtrate from the bases.

The origin of this acid-soluble humin derived from the filtrate from the bases may be due to one specific amino acid. It is known that ferric chloride oxidizes phenols, so that it is possible that the phenol group in tyrosine has reacted here.

The ammonia nitrogen fraction is increased when fibrin is hydrolyzed in the presence of ferric chloride. This is to be expected since the presence of ferric chloride elevates the boiling point of the liquid during hydrolysis. It has been shown by Henriques and Gjaldbaek (4) and Van Slyke (8) that when hydrolysis is carried out at a high temperature there is a transformation of some amino nitrogen into ammonia, indicating a deamination of some amino acids.

As has been stated above, Gortner (2) has shown that if the weight of carbohydrate material present during protein hydrolysis greatly exceeds the amount of protein, an accurate nitrogen determination cannot be obtained. *We have also shown that an exact determination of the chemical groups in proteins cannot be expected when proteins are hydrolyzed in the presence of ferric chloride.*

These results have an important bearing on the application of Van Slyke's method to soils. They would both be reactive in a soil hydrolysate and thus give erroneous values for nitrogen distribution even if all the nitrogen of the soils were contained in the form of protein. If we add to this the fact that non-protein nitrogenous materials are present, it is evident that the nitrogen distribution obtained on a soil hydrolysate has no relationship to those obtained on pure protein material.

SUMMARY

1. When a protein is hydrolyzed in the presence of ferric chloride an accurate nitrogen distribution cannot be obtained.

2. There is a substantial increase in the ammonia nitrogen when hydrolysis is carried out in the presence of ferric chloride. This is due to deamination of some amino acids at the temperature of hydrolysis.

3. The acid-soluble humin nitrogen increases at the expense of a corresponding loss in the filtrate from the bases.

4. This investigation indicates that the earlier conclusion in regard to humin nitrogen precipitated by calcium hydroxide is incorrect. A part of this acid-soluble humin is of protein origin instead of being largely non-protein.

5. This study emphasizes the fact that much of the recent work on the organic nitrogen distribution in soils by the Van Slyke method is entirely unreliable. The data cannot in any way represent the distribution of protein nitrogen in the soil.

It is the intention of one of us to test the suggestion experimentally in regard to the origin of the acid-soluble humin nitrogen as soon as possible.

REFERENCES

(1) GORTNER, R. A. 1913 Studies on the chemistry of embryonic growth. I. Certain changes in the nitrogen ratios in developing trout eggs. *In* Jour. Amer. Chem. Soc., v. 35, p. 632–645.

(2) GORTNER, R. A. 1916 The origin of the humin formed by the acid hydrolysis of proteins. II. Hydrolysis in the presence of carbohydrates and of aldehydes. *In* Jour. Biol. Chem., v. 26, p. 177–204.

(3) GORTNER, R. A., AND HOLM, G. E. 1917 On the origin of the humin formed by the acid hydrolysis of proteins. III. Hydrolysis in the presence of aldehydes. II. Hydrolysis in the presence of formaldehyde. *In* Jour. Amer. Chem. Soc., v. 39, p. 2477–2501.

(4) HENRIQUES, V., AND GJALDBAEK, J. R. 1910 Über quantitative Bestimmung der in Proteine oder in dessen Abbauprodukten vorhanden Peptidbindungen. *In* Ztschr. Physiol. Chem., Bd. 67, p. 8–27.

(5) MORROW, C. A., AND GORTNER, R. A. 1917 The organic matter of the soil: V. A study of the nitrogen distribution in different soil types. *In* Soil Sci., v. 3, p. 297–331.

(6) VAN SLYKE, D. D. 1911 The analysis of proteins by determination of the chemical groups characteristic of the different amino acids. *In* Jour. Biol. Chem., v. 10, p. 15–55.

(7) VAN SLYKE, D. D. 1912 The quantitative determination of aliphatic amino groups: II. *In* Jour. Biol. Chem., v. 12, p. 275–284.

(8) VAN SLYKE, D. D. 1912 The conditions for complete hydrolysis of proteins. *In* Jour. Biol. Chem., v. 12, p. 295–299.

SOIL ACIDITY: I. ITS RELATION TO THE GROWTH OF PLANTS

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It has been known for a long time that certain species of plants are affected unfavorably by soil acidity or what is sometimes called a lack of lime, while in this respect other species are indifferent or may even be affected favorably by an acid soil condition or one of low lime supply. On this basis Contejean (5) made a botanical classification of plants under three general heads: viz., the calciphile, or lime-loving, the calcifuge, or lime-avoiding, and the indifferent. While this system of classification may be valuable from a botanical standpoint, it is not adapted for use in a detailed consideration of the relation of soil acidity and lime to our common agricultural plants, for with a few exceptions all of these plants grow best on soils well supplied with lime. This point is further emphasized by Hilgard (22) who has frequently called attention to the adage,—"A limestone country is a rich country," and also the statement of Hall (17) that any soil containing less than 1 per cent of calcium carbonate will be benefited by liming. The beneficial effects of liming soils on agricultural plants are often due partly to indirect actions of the lime, in which case the use of the term calciphile, or lime-loving, might carry the wrong impression as to the mode of action of the lime.

In this connection it is interesting to note that recent experiments by Pipal (33) of the Indiana Station and White (47) of the Pennsylvania Station show that sorrel (*Rumex Acetosella*) which is commonly supposed to be a lime-avoiding plant is really benefited by liming. As they and also Frear (7) and others have indicated, sorrel is usually found growing on very acid soils because there it meets with the least competition from other weeds and also field crops. It would be found growing even better on limed soils were it not that here other more vigorous plants grow so well that they crowd out the sorrel. Undoubtedly, competition working in conjunction with soil acidity or alkalinity in the way just indicated has a marked influence on the powers of different species of plants for establishing and maintaining them-

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selves under natural conditions (18) and thus has affected the character of the native vegetation found in many regions.

Another method of classification of plants, especially the economic ones, which has been used to some extent in this connection is the division into acid-tolerant, acid-intolerant and indifferent. This method of classification has some desirable features as well as some undesirable ones. It is liable to carry the impression that the acidity of the soil actually attacks the tissues of the plant roots, which, as will be indicated later, is undoubtedly seldom the case. Moreover, it tends to minimize or detract from the importance of factors which operate in an indirect way, and which are sometimes the controlling ones.

The subject of the relation of soil acidity to the growth of plants is really so complex that it seems almost impossible to get a simple system of classification of economic agricultural plants that may be adapted for general use in this connection. The detailed consideration of the harmful effects of soil acidity which follows will bring this out more clearly and forcibly. A considerable number of reasons have been suggested or put forward at various times as to the way in which soil acidity injuriously affects the growth of plants. These may be classified under the following heads:

A. The indirect and general influence of soil acidity on plant growth due to its effect on:
   1. The general fertility of the soil.
   2. The prevalence of plant diseases.
   3. The competitive powers of different species of plants.

B. The direct and specific influence of soil acidity on plant growth due to its effect on:
   1. The supply of available calcium needed by plants as direct plant-food material.
   2. The symbiotic nitrogen-fixing bacteria of the legumes.
   3. The root tissues of plants.

A. THE INDIRECT INFLUENCE OF SOIL ACIDITY ON PLANT GROWTH

1. Effect on the general fertility

It is generally recognized that soil acidity has marked unfavorable influences on the general fertility of soils. These unfavorable influences are due to the effects of soil acidity on the physical, biological and chemical conditions and processes of soils.

The physical condition of clay soils, especially, is generally known to be affected unfavorably by an acid condition, due to an insufficient supply of calcium bicarbonate in the soil solution to keep the clay particles flocculated (16), which condition is necessary for the highly desirable granular or crumb structure. In sandy soils the presence of lime carbonate probably improves the physical condition by acting as a binding agent. In improving the physical condition of soils in these ways, the presence of lime carbonate thus indirectly helps in the proper regulation of the air and moisture supply of soils, which, in turn, is the fundamental basis for developing the proper biological and chemical conditions of soils.

It is fully recognized at present that for nearly all crops an acid soil condition is unfavorable to the highest development of the desirable biological conditions and processes of soils. Nitrification and all forms of nitrogen fixation are often markedly checked by an acid condition. Soil acidity by checking these processes acts as a preservative of the organic matter of soils, and thus prevents or delays the liberation of the plant-food elements bound up in the organic matter itself, as well as the elements in the mineral matter which would be made available by the action of the products of decomposing organic matter. Toxic organic substances may also be preserved (43) and thus tend to accumulate to a harmful degree in acid soils much more than in the non-acid ones.

By direct chemical reactions, as Sykora and the writer (43) have shown, lime carbonate may precipitate and thus inhibit the action of substances toxic to plants. In fact nearly all the chemical reactions which take place in soils are affected unfavorably in regard to fertility by an acid condition.

Under an acid soil condition, the soil phosphorus in particular eventually appears to become less available (38) as a result principally of two reasons: viz., (a)the gradual conversion of the soil phosphorus from calcium phosphate into iron and aluminum phosphates, in which condition the phosphorus becomes with time less and less soluble (42) in the soil solution; and (b) the formation of physical and perhaps chemical complexes of the phosphates with the acidic organic matter (31), in which condition the phosphorus is of low availability because of the very slow rate of decomposition, under acid conditions, of the organic matter, which serves as a protective layer physically and chemically. The addition of lime sometimes retards the immediate effectiveness of insoluble phosphate fertilizers (42), but this is undoubtedly only a temporary condition.[2]

Not only the phosphorus but practically all the soil elements are probably affected as to solubility and availability by the reaction of the soil, whether acid or alkaline. From a purely chemical standpoint it would seem that some elements, namely, calcium, magnesium, sodium and potassium would become less available when the soil becomes acid, while a second group of elements, namely, iron, aluminum, manganese, copper, zinc and other heavy metals, would become more soluble and available. The elements of the first group, especially calcium, constitute the great regulators of the reaction of the soil solution. There are three main reasons why the availability of the first group of elements is usually lowered when a soil becomes acid: viz., (a) the supply of these elements in the form of various compounds becomes less as soil acidity develops; (b) the soil solution must compete for these elements with the insoluble soil acids causing soil acidity; and (c) the possibili-

[2] The addition of lime to acid soils sometimes retards the growth of immediately following crops which have low lime needs, like oats, wheat and corn. This is due to temporary conditions which soon disappear and the lime then greatly benefits these crops as well as those with high lime needs.

ties of solution of these elements are lessened by a lowering of the biological activities which results from soil acidity.

A lowering in availability and concentration in the soil solution of the first-named group of elements in the form of carbonates, especially calcium, is favorable to an increase in solubility and concentration in the soil solution of the second-named group of elements. This is due to the fact that the carbonates of the first group, especially calcium, act as precipitating agents for the elements of the second-named group which might otherwise be in solution in the form of various soluble salts. Soil acidity is thus favorable to a high solubility in the soil solution of the second-named group of elements.

Soil acidity may thus in some cases indirectly affect fertility by limiting the availability of the potassium. Besides neutralizing soil acids which otherwise compete with the soil solution for potassium, the addition of lime may possible in some cases, as has been often assumed, displace potassium in insoluble compounds, and thus make it more available (30), although the recent investigations of Briggs and Breazeale (4) and also others do not support this assumption.

The addition of lime may also make the magnesium more available in the same ways as potassium. This addition of lime to acid soils may in some special cases, where the percentage of magnesia much exceeds that of lime, result in a more favorable lime-magnesia ratio. It is to be noted, however, that an acid soil condition probably lowers the solubility of the magnesia in the soil solution much more than of the calcium oxide, because of the much greater affinity of magnesia (27) for the insoluble soil acids. This is further substantiated by the fact that in most cases dolomitic limestone gives good results when used on acid soils.

With respect to availability or solubility, sodium is affected in practically the same way as potassium, but as regards fertility this is of little importance.

Of the group of elements whose concentration in the soil solution is decreased (29) by the addition of lime to acid soils, iron is one of the most important. This decrease is desirable in that it lessens the chances for the formation of iron phosphate. However, on the other hand, in certain cases an excess of lime may lower the concentration of iron in the soil solution, or the permeability of the root hairs, so much that certain plants like the pineapple (13, 14, 24) and certain varieties of the lupine (32) actually suffer from a lack of iron and become chlorotic, although other plants are not seriously affected unfavorably.

Considerable amounts of manganese have been noted by the writer in the soil solution of certain acid soils. When lime was added to these soils the manganese was not found in any considerable amounts in the soil solution because of the fact that an alkaline condition is favorable for its precipitation. As the writer (42) has previously indicated, this high manganese content of the soil solution of acid soils may possibly in some cases act injuriously on certain plants, because of a high absorption of this element. This idea is

supported by the investigations of White (47) which show that the addition of lime to acid soils lowers the manganese content of clover and sorrel grown thereon. The investigations of Wilcox, Kelly (48) and Johnson (24) emphasize the importance of these manganese relations.

An acid condition is also favorable for the formation of soluble aluminum salts (29) which are believed to be toxic (34) to plants. Hartwell and Pember (20) place considerable emphasis on this indirect effect of soil acidity. The investigations of the Indiana Station (1) on an acid, peaty soil indicated that aluminum nitrate was present in sufficient amounts in the soil solution of this soil to be decidedly toxic to plants. The addition of lime resulted in a precipitation of the aluminum and a prevention of the toxic action. The use of ammonium sulfate without lime may result in the formation of considerable aluminum sulfate which is toxic to plants. Cases of soil acidity in which soluble aluminum salts accumulate in seriously toxic amounts are, however, probably quite exceptional. Acid peaty and sandy soils and others that have received considerable ammonium sulfate or other sulfates and chlorides are most liable to develop a toxic condition in this respect as a result of the solubility of aluminum under these conditions. Silt loams and clays, because of the high "buffer"[3] effects of their large amounts of colloidal matter, undoubtedly largely overcome toxic influences of this kind.

Copper, zinc and other heavy metals if present in the soil in appreciable amounts, either naturally or as a result of their addition in various ways, may become soluble and toxic especially under acid conditions.

As has been shown by Sykora and the writer (43) the addition of lime prevents the toxic action of soluble copper salts. This is probably effected by precipitation of the copper as the insoluble carbonate. Undoubtedly the toxic effects in acid soils of zinc, lead and some others of the heavy metals may be prevented by liming in the same way as with copper. This relation of acidity and lime to possible toxic effects of strong mineral acids and heavy metals is probably a point of considerable importance in certain mining regions where smelter fumes or other waste materials are brought to the soil or where soils have received considerable amounts of certain fertilizers, soil amendments or spray materials.

2. *Effect on prevalence of plant diseases*

In this connection it is important to note that certain fungous diseases like "finger-and-toe" develop to a harmful extent only in acid soils, while other plant diseases like the potato scab are most serious in recently limed soils, and hence at least slight acidity is desirable under many conditions for potato growing. Aside from the cases known to have a direct relation to

[3] The usual meaning of the term "buffer" is given in a footnote page 177. It is used here in a general sense as applying not only to H-ion concentration, but to the concentration of all the constituents in the soil solution.

plant diseases, there are a few special plants like the cranberry, blackberry and water-melon (19) for which a soil of at least slight acidity seems desirable and in some cases necessary for the best growth. It is not known just why these plants grow better on an acid soil, but it seems possible that in some cases plant diseases or mal-nutrition due to a lack of iron where lime is added may again be factors.

3. Effect on plant competition

The influence of soil acidity or alkalinity on the competitive powers of different species of plants for establishing themselves and crowding out others has already been discussed and its method of operation explained. Under native conditions or in case land is left in meadow or pasture for a considerable number of years (18) this influence may be a factor of considerable importance. However, under most farming conditions, which are the primary concern of this paper, this form of competition is largely eliminated by cultivation, and hence need not receive further consideration in this discussion.

General considerations

While it is thus possible to find a few (comparatively very few) cultivated plants that for direct or indirect reasons actually grow better on acid soils than on neutral or alkaline ones, yet it is most important to note that nearly all of our important agricultural plants grow best on neutral or alkaline soils. Reasons have been given why the general fertility of soils is affected favorably by the use of lime on acid soils, but in perhaps 95 per cent of the actual cases of soil acidity there is a vastly more important specific influence on certain plants and there always remains the important question,—why are some of the important agricultural crops affected very much more than others by an acid condition and benefited so much more than the others by the use of lime? For example, why is alfalfa affected more by an acid condition than is oats, or even medium red clover, and why are sugar beets affected much more than potatoes? Apparently these very important differences are not fully explained by the considerations already given regarding the general fertility of the soil, plant competition or plant diseases. In order, therefore, to explain these differences a number of possible *direct* and *specific* influences of soil acidity as already outlined have been suggested at various times. These will now be considered.

B. THE DIRECT AND SPECIFIC INFLUENCE OF SOIL ACIDITY

1. Effect on availability of calcium

The influence of soil acidity on the supply of available calcium which is needed by plants as direct plant-food material has been suggested to explain

at least in part the question under consideration. Brief reference to this has already been made under the head of general fertility, but because of the special *direct* relations of certain plants it is now taken up again and considered more fully. As has been explained, when soils become acid the supply of available calcium in all forms becomes less. That the supply of calcium as plant-food material is not often the limiting factor in acid soils is shown by the fact that the addition of calcium sulfate (19) does not supply the deficiency of these acid soils. Calcium enters the tissues of plants as a necessary constitutent or integral part to only a very small extent. Large amounts are found, however, in many plants in combination with organic acids which are probably by-products of vital life processes. Only calcium in the form of the carbonate or in forms that pass over to the carbonate in the soil meet this special large requirement of certain plants, and hence the calcium as the carbonate seems to be needed directly by these plants for other uses than as plant-food material. These other uses are discussed in detail beginning on page 179.

2. *Effect on symbiotic nitrogen-fixing bacteria*

Since some of the principal legumes are usually greatly benefited by the liming of acid soils, the statement is commonly made that these legumes are sensitive to acidity or are acid intolerant because the nitrogen-fixing bacteria of these legumes are unable to do their work satisfactorily under acid soil conditions. The fact that certain non-legumes are just as injuriously affected by acid soil conditions as certain legumes is evidence that the action of these acid conditions need not necessarily be directly on the legume bacteria which live in the nodules.

In regard to this matter Wheeler (46) has pointed out the fallacy of the common notion in the following words: "On account of a lack of sufficient appreciation of these conditions, the agricultural press and even scientific publications often contain statements to the effect that legumes are in great need of liming, in order that they may develop root nodules and properly assimilate atmospheric nitrogen. Nevertheless the Southern cowpea, serradella, and certain of the lupines are likely to be injured by heavy liming."

Since the legume bacteria live actively in the nodules, the normal medium of these bacteria would seem to be one which is acid to a certain degree, inasmuch as the nodules are in direct connection with the rest of the plant whose circulatory sap is known to be acid. Contrary to the common statement that these legume bacteria require an alkaline medium for active development, it thus appears that they thrive in a medium which is acid to a certain degree, and as the data of Fred and Graul (8) show, even the alfalfa bacteria fix nitrogen to a considerable extent in some acid soils.

There is, however, a time when an acid condition of the soil directly affects the legume bacteria and that is while they are still in the soil prior to the

symbiotic relationship with the plant. The writer has noticed that in case a soil is quite acid, the acidity seems to prevent or delay the plants in becoming infected with the bacteria even though the soil has been thoroughly inoculated with the proper bacteria. Fred and Loomis (10) have shown that when the acidity of media reaches a certain degree the multiplication of alfalfa bacteria is greatly lessened, and hence it is reasonable to believe that an acid condition of the soil affects the acivity, multiplication, rate of movement and hence distribution of the legume bacteria in the soil itself in such a way that the chances for infection of the plant are greatly lessened. This may be due partially to the effects of an acid condition in retarding the neutralization and decomposition of acidic and toxic products, and on the nature and supply of available phosphates (39) and other food material. Phosphates (9) especially are known to have a decided effect on bacterial activity. The supply of calcium carbonate and bicarbonate in the soil solution is generally known to affect greatly the activity of the soil bacteria. This supply is directly affected by an acid condition of the soil.

Frequently in pot cultures of alfalfa on acid soils it happens that at first only one or a few plants become infected even though the soil had been thoroughly inoculated. These plants that become infected grow much more rapidly and become much darker in color than the uninfected ones indicating that if the legume bacteria once gain entrance to the plant, they are able to fix nitrogen fairly energetically even though the soil is quite acid.

The specific influence of soil acidity on the growth of plants thus does not appear to be due to a direct action on the symbiotic nitrogen-fixing bacteria in the nodules.

3. *Effect on the root tissues of plants*

The injurious influence of soil acidity on the growth of certain plants is sometimes assumed to be due to the toxic or destructive effect of the soil acids on the root tissues of the plants. The following statement taken from a well known textbook (26) of soils illustrates this point:

> Free acids are toxic to most agricultural plants. Some plants are much more sensitive than others. Alfalfa, for example, should have a slightly alkaline medium for its best growth, and any acid is very injurious.

Why an acid should be more toxic or destructive to alfalfa roots than to clover roots, or to barley roots than to oats roots, is certainly neither easily explained nor plausible from a chemical or physiological standpoint. Since the sap of practically all plants is acid, and often more acid than the soil solution of the common acid soils, the fallacy of the contention that soil acids often directly affect the root tissues at once becomes apparent. The consideration which immediately follows brings this out clearly.

In an investigation of the acidity[4] (H-ion concentration) of the plant sap of a number of agricultural plants, M. R. Meacham[5] and the writer found that this varies at least between the H-ion exponents of 4.0 and 6.0, or in other terms between the acidities represented by 1/10,000 N and 1/1,000,000 N solutions. The addition of lime to acid soils usually caused a slight lowering in the acidities of the saps of plants grown on these soils. Determination by N. E. Loomis[5] and the writer of the H-ion concentrations of soil extracts from a considerable number of the common cultivated acid soils of Wisconsin of varying degrees of acidity gave a range of H-ion exponents of 4.5 to 8.0. Similarly, in an investigation of a considerable number of acid and alkaline soils, Gillespie (15) found a range of H-ion exponents of 4.4 to 8.6, and Sharp and Hoagland (35) a range of 3.7 to 9.7. It is thus apparent that the acidity of the plant saps is of the same order as that of the water extracts of acid soils, and hence it is not to be expected that the acidity of the soil solution often becomes high enough to be directly seriously toxic or destructive to the plant roots.

The similar and in most cases restricted range of H-ion concentrations of plant saps and soil solutions indicates that similar processes are probably at work in the two cases because of certain analogous conditions which exercise a regulatory function in this respect. Since proteins are *amphoteric*[6] and exert a tremendous *buffer*[7] effect on solutions which bathe them, it is readily explained why the sap of most plants has a rather restricted range of H-ion concentrations. Similarly, the soil constituents, especially the colloidal

[4] The reader is reminded that acidity expressed in terms of H-ion concentration refers only to what is sometimes called "true" acidity resulting from the ionized part of the acids. In many chemical processes and especially in biological acitivity, it has been shown that the effects of acidity are usually due largely to the ionized acidity of H-ions. This does not, however, preclude marked effects from un-ionized acidity in certain cases. The reader is also reminded that Sorensen's (37) method of expressing H-ion concentrations by means of H-ion exponents which is used here is interpreted as follows:

The exponent expressed is the negative exponent or power to which 10 must be raised in order to give the H-ion concentration in grams per liter. Thus:—

Exponent 1.0 means 10^{-1} or $\frac{1}{10}$ gm. H-ions per liter or $\frac{1}{10}$ N acid.

Exponent 5.0 means 10^{-5} or $\frac{1}{100,000}$ gm. H-ions per liter or $\frac{1}{100,000}$ N acid.

An exponent of 7.0 which is that of pure water designates neutrality while one greater than 7.0 designates alkalinity, and one less than 7.0 acidity. The character P_H. proposed by Sorensen is sometimes used to designate the expression—H-ion exponent, or power.

[5] A detailed account of this work will be given in the succeeding numbers of this series of articles on Soil Acidity.

[6] The term "amphoteric" applies to substances which like the proteins may act as either acids or bases to indicators or the surrounding solution. Colloidal soil constituents like organic matter, acid silicates, kaolin, phosphates, iron and aluminum salts and oxides and many others are amphoteric to the soil solution. As the soil solution tends to become more acid these amphoteric substances prevent any rapid change by combining with the acid and also by giving up base to the solution.

[7] The term "buffer" refers to substances which tend to preserve the original H-ion concentration. Salts in solution as well as colloidal matter may act as buffers.

organic and inorganic matter, exert a tremendous buffer effect on the H-ion concentration of the soil solution and in this way restrict and regulate this concentration or acidity to such an extent that the soil solution is usually not acid enough to be directly toxic or destructive to plant root tissues. As the writer (41) has previously pointed out the action of other plant toxins is also inhibited in this way.

The work of Hartwell and Pember (21) of the Rhode Island Station indicates that an acidity of 1/5000 N, HCl, or less, which it is to be noted is more acid than the water extracts of most very strongly acid soils, has no apparent injury on oats, rye, wheat and barley seedlings. These investigators also observed that although barley gains more than rye through the addition of lime to acid soils, yet it is not more sensitive directly to acidity, although it does appear to be to aluminum (20) salts.

Gedroitz (12) also made investigations along this line with water cultures and found the following order of acid resistance beginning with the least resistant: flax, mustard, vetch and barley. Thus barley, although less sensitive directly to acidity, usually responds more to liming than flax, again indicating that the so-called acid intolerance is not due to a direct action of the soil acids on the plants. The acidities which the different plants withstood without injury varied from 1/1000 N to 1/5000 N solutions.

In carefully controlled experiments, Hoagland (23) recently found that a culture having an acidity expressed by an H-ion exponent of 5.2 is very favorable for the growth of barley, and he states that it would appear from these experiments that acid soils with an H-ion concentration of 0.8×10^{-5} (H-ion exponent of 5.1) or less would not be injurious to plants of the cereal group because of excessive acidity.

In certain *unusual* cases of very strongly acid soils, especially peats and sands, it is possible that the acidity of the soil solution may be high enough to affect the plant directly to a serious extent. The writer has on several occasions noted a depression in growth following the addition of soluble fertilizer salts to an unlimed, strongly acid soil. The addition of a salt like K_2SO_4 to an acid soil results, of course, in an increased acidity of the soil solution due to the partial removal of the base by the insoluble soil acids and the liberation of an equivalent amount of free sulfuric acid, or acid sulfate, which being soluble, exerts a much stronger action on the plant than the original insoluble soil acids, and may also increase the soluble aluminum or other salts to a toxic concentration.

It should be noted in this connection that the soil solution in contact with growing plant roots is continually drawn on for nitrates, phosphates and sulfates and receives in return carbonic acid. This not only lowers the possible concentration of soluble mineral acids in the immediate region of the absorbing roots, but the carbonic acid, together with that liberated from decomposing organic matter and biological life, acts on the soil silicates and forms a certain amount of carbonates which undoubtedly have a regulatory influence

on the acidity of the soil solution, especially in the immediate region of the feeding roots, in the same way as they are known to have in other mediums. That this carbonic acid really acts to a considerable extent as a regulator of the acidity of the soil solution of acid soils is indicated by the fact that when even very strongly acid mineral soils are extracted with carbonated water, there is obtained an extract which on boiling is alkaline to phenolphthalein.

General considerations

In most of the common cases of soil acidity it thus appears that the specific harmful influence of the acidity on certain plants is usually not due in any large degree to any of the three suggested reasons which have just been discussed. What appears to the writer to be the most probable way in which this specific influence is exerted will now be discussed and explained in detail.

THE LIME REQUIREMENTS OF PLANTS

Definitions

The designation "lime requirement of soils" has been a common method of speaking of the apparent amount of acidity in soils, but as will be indicated, it is much better to speak of the degree of acidity of soils or acidity directly in some appropriate terms, and use the expression "lime requirement" in reference to plants and not to soils.

The expression "lime requirement of a soil" has usually been understood to mean the amount of lime needed to neutralize the total acidity of the soil, and it has also been quite generally believed that enough lime should be used in practice to neutralize all of this acidity. As the writer (44) has previously indicated, the total acidity of a soil may be very great—much greater than has generally been supposed—and yet in practice it is only necessary to neutralize a small fraction of the total acidity. The amount of lime that can be most profitably used on an acid soil depends not only on the degree of acidity but also on the crop to be grown and the fertility of the soil. Thus the expression "lime requirement of an acid soil" must either have reference to the total acidity of the soil, or if it is to have reference to the amount of lime which should be applied, then, for any particular soil, it is a varying quantity depending on the crop to be grown. As is readily seen, this method of expression leads to a great deal of confusion. Fortunately, this confusion can easily be avoided by adopting a simple method of expressing the degree of acidity of a soil and using the expression "lime requirement" in reference to plants instead of soils.

As the writer (42) has previously indicated, it is usually because the actual lime needs of a plant are not fulfilled that soil acidity exerts a specific injurious influence on certain plants, and not because the acidity is directly toxic or

destructive. Lime is needed by plants for at least two purposes: viz., (1) to furnish the comparatively small amounts of calcium needed as an indispensable constituent of plant substances, in which case it is regarded as a plant-food material; and (2) to act as a neutralizing and precipitating agent of the acids in the plant sap, which are formed as by-products of vital phenomena. Undoubtedly calcium salts of different kinds are suitable for furnishing the calcium needed for the first purpose, but only the carbonate or bicarbonate are suitable for the second purpose. It is undoubtedly for this second purpose that a large part of the calcium found in plants is used, especially in the case of plants which require exceptionally large amounts of calcium.

The expression "lime requirement of a plant" has been used in a general way, but because of insufficient knowledge regarding the relation of soil acidity and liming to plants it has not been clearly defined. It has often been used in referring in a general way to the relation of plant growth to the lime supply of the soil. In order to avoid confusion as to what is meant in the discussions that follow, the writer gives the following definition: *The expression "lime requirement of a plant" refers to the actual lime needs of the plant itself, especially as to the ease and rate at which lime must be secured from the soil by the plant for normal growth.* Thus if a plant has a high lime requirement, then the solution and delivery must be easy and rapid in order to meet the needs of the plant. If a plant has a low lime requirement, then the rate of solution and delivery need also be only low. As is easily seen the rate at which lime must be secured is determined by the lime content and rate of growth of the plant, and the ease with which it must be secured is determined by the feeding power of the plant for lime. The factors which determine the lime requirement may thus be outlined as follows:

1. Lime content of the plant.
2. Rate of growth of the plant.
3. Feeding power of the plant for lime.
   - *a.* Extent of root system.
   - *b.* Character of root system.
   - *c.* Internal acidity of roots.
   - *d.* Excretion of carbonic acid.

It is to be clearly noted that not only the lime content but also the rate of growth and feeding power of a plant are factors that determine the lime requirement of a plant. The feeding power of a plant for lime depends largely on the extent and character of the root system. As is well known, the oat plant has a much more extensive root system than the barley plant and hence is also a much more vigorous feeder. Plants with fibrous and branching root systems that extend and spread fully through the cultivated layer usually have a much stronger feeding power than the tap-rooted plants. Many varieties of alfalfa are tap rooted, and as is well known their rather low feed-

ing power compared with their rate of growth is reflected in the high fertility required for their successful growth. The clovers and some varieties of alfalfa, like Grimm, have a more profusely branched root system which enables them to be grown successfully on poorer and more acid soil. The deep tap-rooted plants, of course, withstand drought better.

Another point in this connection which should be clearly noted is the difference in absorbing or feeding powers of new and old roots. It is at or near the growing regions of the roots where most of the feeding takes place, and hence a plant, of which the cowpea is an excellent example, which grows rapidly and soon produces a fair root system, has a strong feeding power. A comparatively new and hence very active root system of this kind may have a stronger feeding power than a more extensive but older, hardened and less active system. It is probably for this reason that the cowpea is a much stronger feeder than medium red clover. The ability of a plant to grow rapidly on a poor soil is good evidence of a strong feeding power, and conversely, if a plant requires a very rich soil a weak feeding power is indicated.

The investigations of Meacham and the writer previously referred to show that the internal acidity of some plants is considerably higher than of others. Buckwheat, which has a very high feeding power, as is shown by its ability to grow on poor soils, has a very high internal acidity which probably accounts at least partly for this high feeding power in spite of the rather restricted root system. A high internal acidity undoubtedly makes possible a membrane which is more permeable to calcium that is presented as the bicarbonate and also expedites the progressive passage of the calcium salt within the plant. The internal acidity of a plant may thus affect the feeding power even though appreciable amounts of the acids other than carbonic are not excreted by the roots. Carbonic acid is, as is well known, excreted in large amounts by all plant roots.

If these principles regarding the feeding powers of plants are now applied, it becomes evident that if a plant has a high lime content, grows rapidly and has a rather low or medium feeding power, then its lime requirement is very high. Of this, the alfalfa plant is an exceptionally good example. If a plant has a fairly high lime content, grows rapidly, but an exceptionally strong feeding power, then its lime requirement is only medium or low, as is the case with the cowpea and buckwheat. If a plant has a low lime content, a rapid rate of growth but an exceptionally strong feeding power, then its lime requirement is very low, as is the case with oats.

The relation of soil acidity to the lime requirement of plants

As has already been indicated, when mineral soils, even the strongly acid ones, are extracted with carbonated water there are obtained extracts which after boiling are alkaline to phenolphthalein, indicating that in the natural soil solution of even strongly acid mineral soils there is dissolved calcium

bicarbonate, especially in the region of active plant roots where considerable amounts of carbonic acid are produced. Acting on this suggestion, E. Hay[8] and the writer have devised a comparative method for extracting calcium from acid soils with carbonated water. Determinations were made with this method on a considerable number of acid soils and the results correlated with the growths that alfalfa made on these same soils when fertilized but not limed. This correlation revealed a marked parallelism between the growth of the alfalfa and the calcium that was extracted. This indicates that the specific injurious influence of soil acidity on alfalfa and perhaps certain other plants is due to the lack of sufficient calcium as the bicarbonate in the soil solution. In a previous publication the writer (42) pointed this out and explained the mechanism of the reactions involved as follows:

When not disturbed, the soil solution comes to a state of equilibrium with the phosphates, silicates, carbonates, organic compounds and other solid compounds. In case a soil is acid, then solid carbonates in appreciable amounts are usually absent. Hydrolysis and carbonation are the principal processes that bring dissolved mineral substances into the soil solution.

The equilibrium conditions between the carbonic acid in the soil solution and the solid insoluble soil acids and soil silicates may be taken to illustrate the point under discussion. The insoluble soil acids of the acid soil may be represented by H_2X, and $CaSiO_3$ may be taken as a representative silicate. In the soil there are probably present acid silicates which in this system may replace either the soil acids or silicate. In this system of soil acids silicate and carbonic acid, the following reactions are possible.

$$(1)\quad CaSiO_3 + 2H_2CO_3 \rightleftarrows H_2SiO_3 + CaH_2(CO_3)_2$$
$$(2)\quad CaSiO_3 + H_2X \rightleftarrows H_2SiO_3 + CaX$$
$$(3)\quad CaH_2(CO_3)_2 + H_2X \rightleftarrows 2H_2CO_3 + CaX$$

As is evident from these reactions, the concentration of calcium bicarbonate in solution at equilibrium will depend upon, besides the concentration of carbonic acid and temperature, the amount of surface exposed by the calcium silicate and especially upon the amount and *strength* of soil acids present causing soil acidity. If considerable amounts of relatively strong soil acids are present then the concentration and rate of formation and solution of calcium bicarbonate and delivery to the plant will be too low to meet the maximum need of growing alfalfa and certain other plants and hence the growths of these plants will be checked.

The writer has found that the strength, or avidity, of the active soil acids, as well as the amount, is a very important factor in affecting the amount of calcium bicarbonate extracted with carbonated water. The test which the writer (40) has devised for soil acidity gives a collective result called *the degree of acidity* which embodies a measure of both the strength and amount of active acids present in a soil. It is thus especially well adapted in the practical testing of soils.

It is thus evident that the insoluble soil acids are a direct factor in the possibility of a soil meeting the lime requirement of a particular species of plant.

[8] A detailed account of this method and investigation will be given in one of the succeeding numbers of this series of articles on soil acidity.

The relation of soil fertility to the lime requirements of plants

There are many soils in Wisconsin which are medium to strongly acid and yet they are quite fertile. These soils grow fairly good crops of corn, oats, wheat and medium red clover. Some even grow fair crops of alfalfa for at least one or two years, but when such soils are classified it is found that they invariably fall into one of two groups: viz., either (a) they are comparatively new soils which are well supplied with minerals containing the essential elements, or else (b) they are soils which have been heavily fertilized with farm manure. It is to be noted that silicious sandy soils are never found in the first group, and particularly that all soils of at least slight to medium acidity, which have been farmed in a general way for twenty-five years or more with little use of manure, invariably require the addition of lime for the successful production of alfalfa, and often even of red clover. The theory which supposes that the specific effect of soil acidity on plants and bacteria is a toxic or destructive one falls down entirely in explaining this situation, since it does not explain why the same degree of acidity is more injurious in one soil than in another. If the theory which is based on the lime requirements of plants is used, the situation is easily and satisfactorily explained as follows:

It is first important to note that the soils of the two groups which have just been discussed always either have a good supply of calcium silicates and compounds, or they are very active chemically and biologically in the processes that bring calcium bicarbonate into the soil solution, or else they are soils in which there exists a combination of these two conditions. When these soils are extracted with carbonated water, large amounts of calcium as bicarbonate are brought into solution and reference to the mechanism of the reactions involved which have just been described makes it possible to explain why such should be the case. Another point to be emphasized is that acid soils, which are fertile otherwise, quickly produce plants of sufficient vigor to exert their own maximum feeding power for the comparatively insoluble lime compounds. It is now apparent that the reason clover and sometimes even alfalfa grow well on acid soils of the two groups just described is that the plants' lime requirements are satisfied by these soils in spite of the acidity.

The relation of the lime requirements of plants to their degrees of response to liming acid soils

If the theory which is proposed is a tenable one then it should be possible to show that there is a close parallel relation between the lime requirements of plants and their degrees of response to the liming of acid soils, or reciprocally to their ability to grow on acid soils. Unfortunately, there are no data at hand from experiments which were deliberately designed for critically testing this relationship. The data of the Rhode Island Station (19) resulting from extensive investigations by Wheeler, Hartwell and their associates,

are by far the most comprehensive of any dealing with this subject and hence these data have been used a great deal in estimating the relative degrees of response of plants to liming as given in the last column of table 1.

There may be some objections to the use of the Rhode Island data for the present general purpose, because of the considerable amounts of commercial fertilizers which were annually used, and which are known to have a considerable influence on the direct and indirect effects which soil acidity has on plant growth. In the case of the plots which received ammonium sulfate but no lime, the acidity was so abnormally strong that many plants made practically no growth, and hence it is difficult to make reliable comparisons in these cases as to the influence of soil acidity. The other set of unlimed plots, having received sodium nitrate and acid phosphate, are also not especially well adapted for comparison as unlimed plots, since the use of sodium nitrate lowers the acidity and acid phosphate furnishes calcium which under these conditions may be delivered to the plant as calcium carbonate or bicarbonate, and quite largely supply a plant's need in this respect. Because of such reasons as have been given, these data in some cases do not bring out differences which are known to exist from results of other experiments as well as practical farm observations. In the case of the clovers, for example, differences in the response to liming are not indicated, while it seems to be generally believed that alsike will still make a fairly good growth when medium red clover fails because of soil acidity. Undoubtedly plant diseases are sometimes partially the cause of differences in this case. Similarly, crimson clover is also known to grow under more acid conditions than medium red.

In using these data of the Rhode Island Station the aim has been to strike a general average of all the data pertaining to any particular species of plant. In arriving at the final figure for designating the degree of response to liming or the reciprocal figure for ability to grow on acid soils, other scattered available data as well as the writer's own plant house and field observations have been considered and incorporated. The writer therefore regards these figures as being tentative until checked up by experiments which are deliberately planned to test out these points critically.

In arriving at the figures which designate the lime requirements of plants, uncertainties have also arisen in some cases, largely because of the lack of specific data regarding the feeding power of plants for lime. On pages 180 and 181 the factors which influence this feeding power are discussed.

As is indicated in the table the data on the calcium oxide content of plants has been taken quite largely from Wolff's "Aschen Analysen." Wherever possible an attempt was made to secure averages of a considerable number of analyses. This, however, was not always possible, partly because of the desirability of selecting analyses which were made from material cut in the green condition, and in case of a group of plants like the clovers, which are to be critically compared with one another, the necessity of using analyses made on plants which have reached the same stage of maturity. In the

case of alfalfa, the clovers and some other plants, isolated analyses may be found which, relatively, are quite different from those given. This is especially true in case the plants had reached maturity or were cut as hay. It has been conclusively shown that plants may lose a large amount of salts (25), especially lime salts, when allowed to ripen or to be leached after being cut as hay.

Where close comparisons are to be made between similar plants of a group, it is also desirable that the analyses represent plants grown on the same kind of soil and under the same climatic conditions. Because of the great importance of the clovers and alfalfa, attention is called to the fact that the average analyses given for these plants are relatively the same as those reported by Way and Ogston (45), which were made on plants grown under similar conditions.

It is also important to note that the lime content of only the green aerial parts of plants is considered, since it is in these parts that lime is largely needed as an indispensable material for vital phenomena, and hence they give a much better indication of the lime needs of a plant than storage roots, tubers, and woody parts like the trunks of trees.

The relative rate of plant growth refers to the rate of production of green aerial parts in dry weight. In many cases the figures given are simply estimates, which, however can usually be made with considerable confidence.

In the table, abbreviated designations are used as follows:

1 designates – very low
2 designates – low
3 designates – medium
4 designates – high
5 designates – very high

Intermediate stages are designated by halves. In regard to the relative lime contents of plants these designations are used according to the following basis:

1 designates lime content of 0–0.74 per cent, inclusive
1½ designates lime content of 0.75–1.11 per cent, inclusive
2 designates lime content of 1.12–1.49 per cent, inclusive
2½ designates lime content of 1.50–1.86 per cent, inclusive
3 designates lime content of 1.87–2.24 per cent, inclusive
3½ designates lime content of 2.25–2.61 per cent, inclusive
4 designates lime content of 2.62–2.99 per cent, inclusive
4½ designates lime content of 3.00–3.36 per cent, inclusive
5 designates lime content of 3.37 per cent and over.

It is now to be recalled that the lime content of a plant and its rate of growth are two factors, both of which operate in the same direction as regards the lime requirement of a plant. The higher the lime content and the higher the rate of growth the greater will be the lime requirement of the plant. The feeding power of a plant is, however, a factor which works in the opposite

direction to the other two just mentioned. The greater the feeding power of a plant, the other two factors remaining constant, the lower will be the lime requirement of a plant. The lime requirement of a plant is thus determined by three factors, two of which operate in one direction and a third which operates in the opposite direction. The resultant of the three factors indicates the lime requirement.

Before describing the method of obtaining and using this resultant to determine the lime requirement, it is important to note as a fundamental basis, that a high feeding power will entirely offset both a high lime content and a high rate of growth, resulting in a low or medium lime requirement. The cowpea and buckwheat are excellent examples of this. If the lime content, rate of growth, and feeding power are all medium, then, of course, the lime requirement will also be medium and is represented by the figure 3. Using this simple case as a basis for calculation, the designations for lime requirements of other plants may be determined as follows:

If in this same case the factor "feeding power" were lower than 3, the lime requirement would rise above 3 accordingly, and if higher than 3, it would drop below 3 accordingly. The amount of rise or drop is determined by a comparison of the figure which represents the feeding power with a collective figure which represents both the lime content and the rate of growth. This collective figure represents what may be called *the required rate of lime delivery* and being dependent on both lime content and rate of growth is obtained as a relative figure by taking the average of the two. The product of the two is not taken since only relative figures are dealt with.

The method of calculation is best explained and illustrated by giving a few specific examples as follows:

In the case of sweet clover the relative lime content is represented by 3 and the relative rate of growth is estimated at 5, and hence the required rate of lime delivery is represented by 4, which is the average of the two. The feeding power is estimated at $3\frac{1}{2}$, which is lower by $\frac{1}{2}$ than the required rate of lime delivery and hence $\frac{1}{2}$ must be added to the adopted basial figure 3, giving $3\frac{1}{2}$ (medium to high) which represents the lime requirement of sweet clover as obtained by this method of calculation. Similarly, in the case of alsike clover the required rate of lime delivery is found to be 3 and the feeding power is estimated at $3\frac{1}{2}$, which is larger by $\frac{1}{2}$ than the required rate of lime delivery, and hence $\frac{1}{2}$ must be subtracted from the basial figure 3, giving $2\frac{1}{2}$ (low to medium), which represents the lime requirement of alsike clover as obtained by this method of calculation. In a few cases the figures obtained are a little greater than 5 or a little less than 1, but since 5 and 1 are taken to represent the extremes, they are used in these cases in place of the greater or smaller figures. When quarter and three-quarter numbers have resulted from calculations, they have been changed to the half and full numbers, respectively, since such small differences are meaningless when based to a considerable extent on estimates.

If the figures which were obtained in this way, and which are given in the last two columns of table 1, are examined, it is found that there is a remarkably close relationship between the lime requirements of plants and their response to the liming of acid soils, or reciprocally to their ability to grow on acid soils.

A brief discussion of the data relating to the different groups of plants follows:

In the case of the legumes all degrees of lime requirements are found the same as in the case of the non-legumes. The clovers present an interesting group of plants in this respect and the differences in lime content, rate of growth and feeding power for lime adequately explain their differences in lime requirement and ability to grow on acid soils. The same may be said to be true with the other legumes. As is evident, it is entirely unnecessary to postulate that the main effects of soil acidity on legumes are directly traceable to effects on the symbiotic nitrogen-fixing bacteria, or to a destructive or toxic action on the roots.

The cereals present another very interesting group of plants and the higher lime requirement of barley than of the others is also explained.

The hay and pasture grasses are a group of plants whose lime requirements are more similar than of any other group and in many cases it is difficult to estimate their relative feeding power except by resorting to their ability to grow on poor soils, which, however, is quite a reliable index.

Of the fiber crops, cotton and flax are known to have a low lime requirement while that of hemp is about medium. The reasons for this are found in the figures of table 1.

In the miscellaneous crops, various degrees of lime requirements are represented and quite satisfactorily explained.

Most of the garden crops have rather high lime requirements and undoubtedly the more general use of lime for many of these crops would materially cut down the amount of manure required in gardening and truck farming. In the case of a few plants like the carrot and radish, there is a discrepancy of a whole point in the two sets of figures but this may easily be due to lack of data for basing estimates or other special effects of soil acidity. The reason why the cranberry and strawberry grow so well on acid soils is clearly indicated.

There is very little data available regarding the lime content of weeds and only the case of sorrel is given. The reason for its low lime requirement is clearly indicated, although it does have nearly a medium lime content.

A few varieties of trees also are included although data for making estimates are quite meagre. The resulting figures are, however, quite significant, especially in connection with the relation of native tree vegetation to native soil fertility. The trees with the higher lime requirements are found growing on the better soils.

These findings thus substantiate the theory which has been proposed; namely, the main specific injury of soil acidity is that it prevents plants, especially those with high lime requirements and relatively weak feeding powers, from getting the lime from the soil at a sufficiently rapid rate to meet their needs.

Exact concordance between the two columns of figures is of course not to

TABLE 1

The Relation of the Lime Requirements of Plants to their Degree of Response to the Liming of Acid Soils, or Reciprocally to their Ability to grow on Acid Soils

KIND OF PLANT	CALCIUM OXIDE OR LIME IN MATERIAL ANAYZED					RELATIVE RATE OF PLANT GROWTH	REQUIRED RATE OF LIME DELIVERY	FEEDING POWER OF PLANT FOR LIME	LIME REQUIREMENT OF PLANT	RESPONSE OF PLANT TO LIMING, OR RECIPROCALLY ABILITY TO GROW ON ACID SOILS
	Character of material analyzed	Number of analyses	Reported by	Per cent (dry basis)	Relative amount					
I. LEGUMES										
Alfalfa, *Medicago sativa*	Plant in bloom	12	(49)	3.00	4½	4½	4½	2	5	5
Clover, sweet, *Melilotus alba*	Hay	1	(2)	1.94	3	5	4	3½	3½	4
Clover red, *Trifolium pratense*	Plant in bloom	113	(49)	2.40	3½	4	4	3½	3½	3½
Clover, mammoth, *Trifolium medium*	Plant in bloom	3	(49)	2.09	3	4	3½	3½	3	3
Clover, alsike, *Trifolium hybridum*	Plant in bloom	3	(49)	1.62	2½	3½	3	3½	2½	2½
Clover, crimson, *Trifolium incarnatum*	Plant in bloom	4	(49)	1.92	3	3½	3½	4	2½	2
Clover, white, *Trifolium repens*	Plant in bloom	4	(49)	2.21	3	3	3	4	2	2
Clover, Japanese, *Lespedeza striata*	Plant in bloom	1	(49)	1.28	2	3	2½	3	2½	2
Beans, *Phaseolus vulgaris*	Vines	4	(49)	1.32	2	3½	3	4	2	2
Cowpea, *Vigna unguiculata*	Hay	1	(2)	2.70	4	4	4	5	2	2
Lupine, blue, *Lupinus hirsitus*	Hay	3	(49)	1.05	1½	4	3	5	1	1½
Peas, garden, *Pisum sativum*	Vines	6	(49)	3.12	4½	3½	4	2½	4½	4
Peanuts, *Arachis hypogaea*	Vines	1	(6)	2.08	3	3	3	3	3	3
Soybean, *Glycine hispida*	Hay	2	(2)	2.76	4	4	4	4	3	3
Serradella *Ornithopus sativus*	Plant in bloom	3	(49)	2.19	3	2	2½	3	2½	2
Vetch, common, *Vicia sativa*	Green plant	25	(49)	1.96	3	3	3	4	2	2½
II. NON-LEGUMES										
a. Cereals										
Barley, *Hordeum sativum*	Before bloom	3	(49)	0.95	1½	4	3	3	3	3½
Buckwheat, *Fagopyrum esculentum*	In bloom	7	(49)	3.33	4½	4½	4½	5	2½	2

Corn, *Zea mays*	In bloom	7	(49)	0.83	1½	5	3½	3½	3	2½
Oats, *Avena sativa*	Before bloom	9	(49)	0.53	1	4	2½	4	1½	1½
Rice, *Oryza sativa*	Entire plant	1	(28)	0.23	1	2	1½	2½	2	2
Rye, *Secale cereale*	Before bloom	1	(49)	0.51	1	4	2½	4	1½	1
Wheat (winter), *Triticum vulgare*	Before bloom	2	(49)	0.48	1	3	2	2½	2½	2
b. Hay and pasture grasses										
Italian rye-grass, *Lolium Italicum*	Hay stage	2	(2)	1.07	1½	3½	2½	4	1½	2
Johnson-grass, *Sorghum halepense*	Hay stage	1	(49)	0.62	1	4	2	4	1	2
Kentucky blue-grass, *Poa pratensis*	Hay stage	4	(49)	0.42	1	3	2	2½	2½	2½
Meadow Fescue, *Festuca pratensis*	Hay stage	4	(2)	0.57	1	3	2	3½	1½	2
Millet (species not stated)	Hay stage	1	(11)	0.46	1	3½	2½	3½	2	2
Orchard-grass, *Dactylis glomerata*	Hay stage	4	(2)	0.46	1	3½	2½	3½	2	2½
Perennial rye-grass, *Lolium perenne*	Hay stage	2	(2)	0.64	1	3	2	3½	1½	2
Redtop, *Agrostis vulgaris*	Hay stage	4	(2)	0.57	1	3	2	3½	1½	1
Timothy, *Phleum pratense*	Hay stage	2	(2)	0.71	1	3½	2½	3½	2	2½
c. Fiber crops										
Cotton, *Gassypium hirsutum*	Entire plant	1	(49)	0.75	1½	3	2½	3½	2	1½
Flax, *Linum usitatissimum*	Entire plant	1	(49)	0.75	1½	1½	1½	1½	1½	1
Hemp, *Cannabis sativa*	Entire plant	1	(49)	0.76	1½	5	3½	3½	3	3
d. Miscellaneous crops										
Hops, *Humulus lupulus*	Entire plant	3	(49)	2.30	3½	3	3½	3	3½	3½
Potato, common *Solanum tuberosum*	Vines	6	(49)	2.80	4	2	3	3	3	2½
	Tubers	59	(49)	0.10						
Potato, sweet, *Ipomoea batatas*	Vines	1	(3)	1.70	2½	2½	2½	3	2½	2
Rape, *Brassica napus*	In bloom	6	(49)	1.78	2½	3½	3	3	3	3½
Sugar beet, *Beta vulgaris*	Leaves	25	(49)	3.00	4½	2½	3½	2	4½	5
	Roots	149	(49)	0.23						
Sorghum, *Sorghum vulgare, saccharatum*	Entire plant	1	(36)	0.26	1	4	2½	4	1½	2
Sugar cane *Saccharum officinarum*	Entire plant	13	(49)	0.19	1	4	2½	3½	2	2
Tobacco, *Nicotiana tabacum*	Entire plant	1	(42)	3.40	5	3	4	2	5	4
Turnip, *Brassica rapa*	Leaves	10	(49)	3.83	5	2	3½	2½	4	3
	Roots	32	(49)	0.85						

TABLE 1—*Continued*

KIND OF PLANT	CALCIUM OXIDE OR LIME IN MATERIAL ANALYZED					RELATIVE RATE OF PLANT GROWTH	REQUIRED RATE OF LIME DELIVERY	FEEDING POWER OF PLANT FOR LIME	LIME REQUIREMENT OF PLANT	RESPONSE OF PLANT TO LIMING OR RECIPROCALLY ABILITY TO GROW ON ACID SOILS
	Character of material analyzed	Number of analyses	Reported by	Per cent (dry basis)	Relative amount					
e. Garden crops, etc.										
Cabbage, *Brassica oleracea, capitata*	Leaves	2	(49)	5.93	5	2½	4	2½	4½	4
	Heart	3	(49)	1.17						
Carrot, *Daucus carota*	Leaves	8	(49)	4.45	5	2½	3½	2	4½	3½
	Root	11	(49)	0.62						
Cauliflower, *Brassica oleracea, botrytis*	Leaves and stalk	1	(49)	2.04	3	2	2½	1	4½	4
Cranberry, *Vaccinium macrocarpon*	Leaves		(2)	0.40	1	1	1	3	1	1
	Fruit		(2)	0.03						
Celery, *Apium graveoleus*	Young shoots	1	(49)	1.88	2½	3	3	1	5	5
Grape, *Vitis*	Leaves	18	(49)	2.55	3½	3½	3½	3	3½	3
Kohl-rabi, *Brassica oleracea, caulo-rapa*	Leaves	1	(49)	4.64	5	2	3½	2½	4	4
Lettuce, *Lactuca sativa*	Head lettuce	3	(49)	2.65	4	3½	4	2½	4½	5
Onion, *Allium cepa*	Leaves	1	(49)	3.63	5	1	3	1½	4½	5
	Bulb	2	(49)	1.21						
Rhubarb, *Rheum rhaponticum*	Leaves	1	(49)	0.32	1	4	2½	3	2½	2½
	Stalks	1	(49)	1.45						
Radish, *Raphanus sativus*	Leaves	1	(49)	4.30	5	3	4	3	4	3
	Root	1	(49)	1.38						
Spinach, *Spinacia oleracea*	Top	2	(49)	1.96	3	3	3	2	4	5
Strawberry, *Fragaria chiloensis*	Vines		(2)	1.22	2	1½	2	3½	1½	2
	Fruit		(2)	0.07						
f. Weeds										
Sheep sorrel, *Rumex acetosella*	Whole plant	1	(49)	1.63	2½	2	2½	4	1½	1
g. Trees										
Chestnut	Leaves	1	(49)	2.25	3½	2½	3	3	3	3
Mulberry	Leaves	1	(49)	5.14	5	2	3½	2	4½	4½
Oak	Leaves	1	(49)	0.91	1½	2½	2	3½	1½	2
Pine	Needles	8	(49)	0.52	1	2	1½	3½	1	1

be expected in all cases, partly because of the meagre data available in some cases for basing conclusions, and partly because of the intervention of other factors of general soil fertility which in some cases may be much greater than others. The writer thus wishes to state clearly that many minor errors surely exist in the figures of table 1, which only careful investigation can eliminate. On the basis of the data available the validity of the theory in the main, however, seems certain.

The foregoing considerations and conclusions are especially important in formulating a simple, practical, and rational system of determining the amount of lime which should be applied under practical farming conditions. It is thus evident that the amount of lime which should be applied, assuming that a good quality is to be used, depends upon the following factors:

1. Lime requirement of crop or crops to be grown.
2. Degree of acidity of the soil.
3. Fertility of the soil.

How these factors may be combined into a single usuable quantity will be discussed in the succeeding articles.

SUMMARY

With a few exceptions agricultural plants grow best on soils well supplied with readily available lime. To be readily available, lime may exist either as the carbonate, as an easily hydrolyzable silicate or salt, or as a constituent of easily decomposable organic matter. The classification of agricultural plants as being lime-loving, lime-avoiding and indifferent, or as being acid-intolerant, acid-tolerant and indifferent, leads to confusion and gives the wrong impression regarding the relation of lime and soil acidity to plant growth. The subject is very complex and, as has been indicated, soil acidity has many indirect and general influences on soil fertility due to its effect on physical, chemical, and biological conditions of the soil.

It is well known that an acid condition is unfavorable to the highest development of desirable physical and biological soil conditions. An acid condition usually lowers the availability of nearly all the essential elements. On the other hand, soil acidity usually favors the accumulation and solubility of toxic organic and inorganic substances. Among these toxic substances soluble aluminum salts have been noted by a number of investigators, and possibly in some cases manganese salts should also be considered. In certain unusual cases of soil acidity sufficient amounts of these toxic substances may be present to be very harmful to some plants. The relation of soil acidity and liming to malnutrition due to a lack of iron in the plant, to plant diseases and to plant competition also need to be considered in a few special cases, particularly in cases where soil acidity appears to be a favorable condition.

Besides the indirect influences which affect all plants, and some probably to a considerable extent, soil acidity has a specific influence which affects

some plants like alfalfa and sugar beets very much more than others, like cowpeas, potatoes, and oats. At various times this specific influence of soil acidity has been ascribed to at least three causes: viz., (a) its effect on the supply of available calcium needed by plants as direct plant-food material, (b) its effect on the symbiotic nitrogen-fixing bacteria, (c) and its toxic or destructive effect on the root tissues of plants.

The supply of available calcium in all forms becomes less as soils become acid, but usually there is still sufficient present to furnish that needed as direct plant-food material. Since the symbiotic nitrogen-fixing bacteria live in the nodules, soil acidity cannot affect them directly, except before they enter into symbiosis, when it may lessen their activity and delay the time of infection. Since the relation of non-legumes to soil acidity runs parallel with the relation of the legumes, it follows that the direct influence is not on the legume bacteria but on the plants themselves.

That this direct effect on the plants is not often due to a destructive action of the acidity on the root tissues is evident from the fact that experiments have shown that plant roots are unaffected by solutions of a higher acidity than that of most acid soil solutions. This is further substantiated by the fact that the acidity (H-ion concentration) of the sap of most plants is of the same order as that of the soil solution of most acid soils, indicating that similar processes are probably at work in the two cases, as a result of certain analogous conditions which exercise a regulatory function in this respect. It is undoubtedly largely proteins in the case of plants, and collodidal organic and inorganic matter, especially silicates, in the case of soils which act as "buffers" and thus bring about this regulation of acidity to a considerable extent, preventing rapid changes and unusually high degrees of soluble acidity.

In most cases it thus appears that the *main* specific harmful influence of soil acidity on certain plants is not due to any of the three suggested reasons, but to its influences in preventing these plants from getting at a sufficiently rapid rate the calcium as the carbonate or bi-carbonate which is needed to neutralize and precipitate certain acids in the plants themselves, which are probably largely by-products, produced as the result of certain vital reactions in the growth of plants. If calcium in these forms is not furnished at a sufficiently rapid rate, then the rate of these reactions is lowered accordingly as is also the rate of plant growth.

Each species of plant has a certain lime requirement which must be satisfied for maximum plant growth and this lime requirement is defined by the writer as follows: *The expression "lime requirement" of a plant refers to the actual lime needs of theplant itself, especially as to the ease and rate at which lime must be secured from the soil by the plant for normal growth.* Thus if a plant has a high lime requirement, then the solution and delivery must be rapid and easy in order to meet the needs of the plant.

The three main factors which determine the lime requirement of a plant

are: (a) lime content, (b) rate of growth, and (c) feeding power of the plant for lime. The first two factors operate in one direction while the third operates in the opposite direction. That is, the higher the lime content and the rate of growth, the higher will be the lime requirement, and conversely. Also, the higher the feeding power for lime, the lower will be the lime requirement, and conversely. The resultant of these three gives the lime requirement of the plant. A simple method of expressing these factors and obtaining the resultant is described in this article.

A table is also given in which are expressed the lime requirements of 62 species of plants as obtained by this method. These lime requirements are compared with corresponding figures which represent the relative response of these plants to the liming of acid soils, or reciprocally to their ability to grow on acid soils. The comparison reveals a close correspondence and hence substantiates the theory which has been proposed that, usually, the main specific injury of soil acidity is that it prevents plants, especially those with high lime requirements and relatively weak feeding powers, from getting the lime from the soil at a sufficiently rapid rate to meet their needs. This is further substantiated by the parallel relation found between the amount of growth of alfalfa on acid soils and the amount of calcium which could be extracted with carbonated water from these soils. These considerations are especially important in formulating a practical system of using lime, especially as regards the amount to be used which, as is discussed in detail, is dependent on the lime-requirement of the crop, the degree of acidity of the soil, and the fertility of the soil.

REFERENCES

(1) Abbott, J. B., Connor, S. D., and Smalley, H. R. 1913 Soil acidity, nitrification, and the toxicity of soluble salts of aluminum. Ind. Agr. Exp. Sta. Bul. 170, p. 329–374.

(2) Beal, W. H. 1889 Compilations of analyses of fodder articles, fruits, sugar, producing plants, dairy products, etc. *In* Mass. State Agr. Exp. Sta. 7th Ann. Rpt. (1889), p. 291–313.

(3) Blair, A. W. 1908 Total ash and lime in some feeding materials and fruits. *In* Fla. Agr. Exp. Sta. Ann. Rpt. 1908, p. 46.

(4) Briggs, L. J., and Breazeale, J. F. 1917 Availability of potash in certain orthoclase-bearing soils as affected by lime or gypsum. *In* Jour. Agr. Res., v. 8, no. 1, p. 21–28.

(5) Contejean 1881 Geographie botanique. Influence du terrain sur la vegetation. 143 p. Paris. (From Hilgard, E. W. (22), p. 521.)

(6) Corbett, L. C. 1907 Peanut. *In* Bailey's Cyclopedia of American Agriculture, v. 2, p. 519.

(7) Frear, W. 1915 Sour soils and liming. Pa. Dept. Agr. Bul. 261, p. 101.

(8) Fred, E. B., and Graul, E. J. 1916 The gain in nitrogen from growth of legumes on acid soils. Wis. Agr. Exp. Sta. Res. Bul. 39, p. 1–42.

(9) Fred, E. B., and Hart, E. B. 1915 The comparative effect of phosphates and sulphates on soil bacteria. Wis. Agr. Exp. Sta. Res. Bul. 35, p. 35–66.

(10) FRED, E. B., AND LOOMIS, N. E. 1917 Influence of hydrogen-ion concentration of medium on the reproduction of alfalfa bacteria. *In* Jour. Bact., v. 2, no. 6, p. 629–633.

(11) FORBES, E. B., BEEGLE, F. M., AND MENSCHING, J. E. 1913 Mineral and organic analyses of foods. Ohio Agr. Exp. Sta. Bul. 255, p. 211–232.

(12) GEDROITZ, K. K. 1910 The effect of acids, alkalis, and some inorganic salts on plants. *In* Zhur. Opuitn. Agron. (Russ. Jour. Exp. Landw.), v. 11, no. 4, p. 544–578, no. 5, p. 641–678.

(13) GILE, P. L. 1911 Relation of calcareous soils to pineapple Chlorosis. Porto Rico Agr. Exp. Sta. Bul. 11, 45 p.

(14) GILE, P. L., AND AGETON, C. N. 1914 The effect of strongly calcareous soils on the growth and ash composition of certain plants. Porto Rico Agr. Exp. Sta. Bul. 16, 45 p.

(15) GILLESPIE, L. J. 1916 The reaction of soil and measurements of hydrogen-ion concentrations. *In* Jour. Wash. Acad. Sci., v. 6, no. 1, p. 8–16.

(16) HALL, A. D. 1908 The Soil, p. 40. E. P. Dutton Co., New York.

(17) HALL, A. D. 1908 Fertilizers and Manures, p. 253. E. P. Dutton Co., New York.

(18) HALL, A. D. 1910 The adaptation of the plant to the soil. *In* Jour. Roy. Hort. Soc. (London), v. 36, p. 1–21.

(19) HARTWELL, B. L., AND DAMON, S. C. 1914 The comparative effect on different kinds of plants of liming an acid soil. R. I. Agr. Exp. Sta., Bul. 160, p. 407–446.

(20) HARTWELL, B. L., AND PEMBER, F. R. 1918 Aluminum as a factor influencing the effect of acid soils on different crops. *In* Jour. Amer. Soc. Agron., v. 10, no. 1, p. 45.

(21) HARTWELL, B. L., AND PEMBER, F. R. 1907 The relation between the effects of liming and of nutrient solutions containing different amounts of acid, upon the growth of certain cereals. *In* R. I. Agr. Exp. Sta. 20th Ann. Rpt. (1907), p, 358–380.

(22) HILGARD, E. W. 1905 Soils, p. 365. Macmillan Co., New York.

(23) HOAGLAND, D. R. 1917 The effect of hydrogen and hydoxyl-ion concentration on the growth of barley seedlings. *In* Soil Sci., v. 3, no. 6, p. 547–560.

(24) JOHNSON, M. O. 1917 Manganese as a cause of the depression of the assimilation of iron by pineapple plants. *In* Jour. Indus. Engin. Chem., v. 9, no. 1, p. 47–49.

(25) LECLERC, J. A., AND BREAZEALE, J. F. 1908 Plant food removed from growing plants by rain or dew. *In* U. S. Dept. Agr. Yearbook 1908, p. 389.

(26) LYON, T. L., FIPPIN, E. O., AND BUCKMAN, H. O. 1916 Soils, Their Properties and Management, p. 537. Macmillan Co., New York.

(27) MACINTIRE, W. H., WILLIS, L. G., AND HARDY, J. I. 1914 The non-existence of magnesium carbonate in humid soils. Tenn. Agr. Exp. Sta. Bul. 107, p. 151–202, 2 figs.

(28) MCDONNELL, C. C. 1901 A chemical investigation of the rice plant and of the product and by-products of the rice industry. S. C. Agr. Exp. Sta. Bul. 59, 16 p.

(29) MORSE, F. W., AND CURRY, B. E. 1908 A study of the reactions between the manurial salts, clay, mucks, and soils. N. H. Agr. Exp. Sta. 19th and 20th Ann Rpts. (1908–09), p. 271–293.

(30) MORSE, F. W., AND CURRY, B. E. 1909 The availability of the soil potash in clay and clay loam soils. N. H. Agr. Exp. Sta. Bul. 142, p. 39–58.

(31) PETERSON, P. P. 1912 Effect of heat and oxidation on the phosphorus of the soil. Wis. Agr. Exp. Sta. Res. Bul. 19, p. 1–16.

(32) PFEIFFER, T., AND BLANCK, E. 1914 Die Kalkfeindlichkeit der Lupine. *In* Mitt. Landw. Inst. Breslau, Bd. 7, No. 2, p. 201–233.

(33) PIPAL, F. J. 1916 Red sorrel and its control. Ind. Agr. Exp. Sta. Bul. 197.

(34) RUPRECHT, R. W. 1915 Toxic effects of iron and aluminum salts on clover seedlings. Mass. Agr. Exp. Sta. Bul. 161, p. 125–129.

(35) SHARP, L. T., AND HOAGLAND, D. R. 1916 Acidity and adsorption in soils as measured by the hydrogen electrode. *In* Jour. Agr. Res., v. 7, no. 3, p. 123–145.

(36) SHOREY, E. C. 1906 The composition of some Hawaiian feeding stuffs. Hawaii Agr. Exp. Sta. Bul. 13, p. 16.

(37) SORENSEN, S. P. L. 1912 Ueber die Messung und Bedeutung der Wasserstoff ionen-koncentration bei biologischen Prozessen. *In* Ergeb. Physiol., v. 12, p. 393–532.

(38) STODDART, C. W. 1909 Soil acidity in its relation to lack of available phosphates. *In* Jour. Indus. Engin. Chem., v. 1, no. 2, p. 69–74.

(39) TRUESDELL, H. W. 1917 The effect of phosphorus on alfalfa and alfalfa bacteria. *In* Soil. Sci., v. 3, no. 1, 77–98 p.

(40) TRUOG, E. 1915 A new test for soil acidity. Wis. Agr. Exp. Sta. Bul. 249, p. 1–16.

(41) TRUOG, E. 1915 Soil acidity and methods for its detection. *In* Science, n.s., v. 42, no. 1084, p. 505–507.

(42) TRUOG, E. 1916 The utilization of phosphates by agricultural crops, including a new theory regarding the feeding power of plants. Wis. Agr. Exp. Sta. Res. Bul. 41, p. 34.

(43) TRUOG, E., AND SYKORA, J. 1917 Soil constituents which inhibit the action of plant toxins. *In* Soil Sci., v. 3, no. 4, p. 333–351.

(44) TRUOG, E. 1917 Letter on the avidity of soil acids. *In* Jour. Indus. Engin. Chem., v. 9, no. 11, p. 1072.

(45) WAY, J. T., AND OGSTON, G. 1870 Ash analyses of plants. *In* Wolff, E. (49), pt. 1, p. 178.

(46) WHEELER, H. J. 1913 Manures and Fertilizers, p. 307. Macmillan Co., New York.

(47) WHITE, J. W. 1916 Concerning the growth and composition of clover and sorrel as influenced by varied amounts of limestone. *In* Pa. Agr. Exp. Sta. Ann. Rpt. 1913–14, p. 46–67.

(48) WILCOX, E. V., AND KELLEY, W. P. 1912 The effect of manganese on pineapple plants. Hawaii Agr. Exp. Sta. Bul. 28, 20 p.

(49) WOLFF, E. 1870 and 1880 Aschen Analysen, pt. 1, 1870, 194 p., pt. 2, 1880, 170 p., Wiegant and Hempe, Berlin.

THE EFFECT OF HEAT ON SOME NITROGENOUS CONSTITUENTS OF SOIL

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In the investigations of the effect of heat on soil the only nitrogenous constituents existing as such which have been determined quantitatively are ammonia and nitrates. Kelley and McGeorge (4) also determined the change in monoamino, diamino and amid nitrogen in the hydrochloric acid hydrolysed, untreated soil and the same soil after heating to 200°C. In this laboratory, methods have been developed for the determination of amino acid, nitrogen and soluble non-protein nitrogen (12, 13). These constituents had previously never been determined in soils by the use of such reagents as precluded their formation from more highly organized material or their partial destruction during the procedure for the determination. A method for the determination of ammonia (11) which gives more nearly absolute results than those usually used also has been worked out. It was because of these and other considerations that the work herein reported was undertaken.

For an extensive article on the literature on soil sterilization, the paper by Kopeloff and Coleman (6) appearing in a recent number of this journal should be consulted. Only the more important investigations having a direct bearing upon the phases of the subject taken up by us will be discussed here.

Richter in 1896 (14) found that heating a soil to 100°C. increased the amount of nitrogen soluble in water and dilute hydrochloric acid, while the water-soluble organic matter was increased about 300 per cent.

Nitrification was found by Deherain and DeMoussy (2) to be inhibited completely in soils heated at 120°C. for 1 hour in the autoclave. Upon inoculation of the heated soil with fresh soil an abnormally large amount of nitrates was produced. Pickering (9, 10) found that heating soils caused an increase in the total soluble organic matter. Upon inoculation of the heated soil and incubation for a few weeks, this excess soluble organic matter was decreased somewhat. He also concluded that the action of the common volatile antiseptics caused about the same chemical change in a soil as heat at 60–75°C.

Russell and his associates (15, 16) have carried out more comprehensive work along these lines than any other investigators. The results of their researches have led them to propose the theory that protozoa are an important limiting factor of bacterial growth. The conclusions to be drawn from their work having direct bearing on the subject matter of this paper, are as

follows. Partial sterilization of the soils, that is, heating at 98°C. or treatment with toluene, caused an increase in the amount of ammonia. This amount of ammonia was further increased to a relatively great extent by incubation of the partially sterilized soil. Nitrates were not changed by heat at 98°C. nor was there any appreciable change after standing in the amount present either in the uninoculated soil or the soil inoculated with a fresh soil. Their contention was that nitrifying organisms would not work in the heated soil. "Unstable nitrogen compounds" were increased by heat at 98°C. to about the same relative extent as ammonia. They were also increased by subsequent incubation of the soil but to a less extent than the ammonia. The "unstable nitrogen compounds" were determined by distillation of the soil with magnesia at 100°C. for an arbitrarily fixed length of time. The amount of ammonia thus formed minus that formed by distillation of the soil at 38°C. was taken to be unstable nitrogen. The amount of humus present in the heated soil was somewhat less than in the unheated.

Lyon and Bizzell (7, 8) studied the changes in soluble matter of soils brought about by steaming at 2 atmospheres and the further changes at the end of various lengths of time after inoculation with fresh and with steamed soils. As they point out, the results obtained on some soils are quite inconsistent when compared with others, but the general conclusions to be drawn from their work are as follows. Steaming invariably increased the amount of soluble organic matter and ammonia and decreased the nitrates. After inoculation with fresh soil and incubation for periods ranging from 5 to 19 weeks, nitrates were often decreased and never increased but to a slight extent. Ammoniacal nitrogen was invariably increased by incubation for periods up to 19 weeks, while total soluble matter was decreased. One sample of steamed soil was inoculated with fresh soil and allowed to incubate for 6½ months. Nitrates were greatly increased, total soluble matter was increased and ammonia was considerably decreased. These results were just opposite to those found in the same soil after incubation for the shorter periods.

Schreiner and associates (17) found that heating at 135°C. in the autoclave for 3 hours increased the amount of xanthine and hypoxanthine when they were found in the untreated soil. When they were not found in the unheated soil appreciable amounts could be separated from the heated soil. Cytosin, guanine and arginine not found in the unheated soil were isolated after heating the soil. Pentosans, histidine, creatinine and dihydroxystearic acid were found to be increased by the treatment in the autoclave. The amount of nucleic acid was lessened.

EXPERIMENTAL

The plan of our work was as follows:

Large samples of four soils were put through a ⅛-inch mesh sieve and each sample thoroughly mixed. Several small samples of each of the soils of 350 gm. each were weighed into 500-

cc. Florence flasks, cotton plugs inserted and then subjected to the following treatments: Two hours at 100°C., dry heat; 2 hours at 200°C., dry heat; the same treatment and then inoculated with fresh soil emulsion and incubated for 10 weeks, the same treatment and then inoculated as above and incubation for 20 weeks; 15 pounds in the autoclave for 3 hours; 10 pounds in the autoclave for 3 hours per day for 3 successive days; the same treatment, inoculated as above and incubated for 10 weeks; the same treatment, inoculated as above and incubated for 20 weeks. All treatments and incubations were carried out in duplicate. Before incubation the soils were made up to the optimum moisture content and maintained at that throughout the incubation period. After the soils had been subjected to the various treatments they, together with duplicate samples of the untreated soils, and the inoculated soils, after they had incubated the specified length of time, were subjected to analysis as described below.

METHODS OF ANALYSIS

Loss on ignition. Approximately 2 gm. of soil were accurately weighed into covered porcelain crucibles. The crucible and contents were then placed in the oven at 100–105°C. for 3 hours and then weighed again. This gave the amount of hygroscopic moisture. These results are not recorded here because of the fact that the soils were not held under uniform conditions between the time of the completion of the respective treatments and the weighing into the crucibles. The determination, however, was necessary, in order to calculate the results to the oven-dry basis. After obtaining the oven-dry weight the crucibles were placed in the muffle furnace and heated for 2 hours or more at a bright red heat. All determinations were made in duplicate.

Nitrate nitrogen. Nitrates were determined by the modification of the aluminum reduction method as proposed by Burgess (1) using the apparatus proposed by us (11).

Ammonia nitrogen. This was determined by the method developed in this laboratory (11).

Amino acid nitrogen. The amino acid nitrogen was determined according to our modification (12) of the Kober (5) method.

Soluble non-protein nitrogen. This group of compounds were determined by the Greenwald (3) method as modified by us for use on soil extracts (13).

DESCRIPTION OF THE SOILS

Calhoun silt loam. This soil was obtained from Van Buren County, Iowa, which is situated in the Southern Iowa loess soil area. It is a fairly fertile soil, acid in reaction and rather low in organic matter.

Miami silt loam I. This was obtained from one of the station orchards which is situated in the Wisconsin drift soil area. This soil is similar to the Calhoun silt loam but contains a little sand, is lighter in color and contains less organic matter.

Miami silt loam II. This comes from a different place in the same orchard

as the above soil. It is darker in color and contains more organic matter than the other sample of Miami silt loam.

Peat soil. This soil was secured from a peat spot in one of the experiment station pastures. It is a typical peat soil, black in color and quite light in weight. The amount of organic matter present is shown below by its loss on ignition.

In table 1 are found the results obtained with the Van Buren County soil. The values for the loss on ignition have been calculated on the basis of the ignited soil. The reason for adopting this standard instead of the oven-dry basis was that some loss of organic material was caused by the more severe

TABLE 1

Calhoun silt loam soil

TREATMENT	LOSS ON IGNITION	TOTAL NITROGEN	PER CENT OF TOTAL NITROGEN							
			Ammonia		Amino acid		Nitrate		Soluble non-protein nitrogen	
			Original basis	Final basis	Original basis	Final basis	Original basis	Final basis	Original basis	Final basis
	per cent	*per cent*								
Air-dry	5 80	0.181	0.60		0.270		0.325		14.45	
100°C.; 2 hours	5.47		0.89		0.230		0.450		16.70	
10 pounds; 9 hours	5.88	0.177	3.01	3.08	0.530	0.54	0.895	0.915	22.80	23.6
15 pounds; 3 hours	5.59	0.179	1.34	1.35	0.430	0.44	0.660	0.670	17.10	17.3
200°C.; 2 hours	4.67	0.175	12.70	13.10	0.890	0.92	0.120	0.120	30.30	31.3
10 pounds; 9 hours; 10 weeks incubation			6.14	6.28	0.415	0.42	2.090	2.130	20.80	21.3
10 pounds; 9 hours; 20 weeks incubation			4.89	5.00	1.150	1.18	2.650	2.700	20.80	21.3
200°C.; 2 hours; 10 weeks incubation			17.05	17.60	0.340	0.35	1.280	1.320	30.60	31.6
200°C.; 2 hours; 20 weeks incubation			17.20	17.80	1.730	1.69	2.320	2.400	32.50	33.6

treatments, and hence it would be incorrect to express the percentages on the oven-dry weight of each soil. It is seen that the percentages of the various nitrogenous constituents are expressed on two bases. The results in the "original basis" columns are expressed in per cent of the total nitrogen originally in the soil. The "final basis" results represent the per cent of the total nitrogen left in the soil. No nitrogen was lost by the 100°-heated soil and it was assumed that no change in the total nitrogen was occasioned by the incubation. This probably is not strictly correct but the change in total nitrogen in the short incubation periods of 10 and 20 weeks would be very slight.

The figures giving the loss on ignition may be taken to represent the amount

of organic matter in the soils. There is seen to be some change in the organic matter of the soil by all the treatments but perhaps the only one great enough to be of any significance is the 200° treatment, and here the change from 5.80 to 4.67 per cent represents a loss of 19.5 per cent of the organic matter.

The next column of the table gives the per cent of nitrogen in the soil from the various treatments. Here none of the treatments show much of a loss. The results for the soil heated at 15 pounds might well be considered to vary from the untreated soil by no more than the experimental error, but the other two figures are probably well without the error. The 200° treatment causes a loss of 3.3 per cent of nitrogen, considerably less than the total loss of organic matter. At 10 pounds, a smaller loss of 2.21 per cent was found.

The results for the changes in the nitrogenous constituents in the incubated soils will be taken up now. The results for the various heat treatments have been plotted and appear in figures 1 to 4, and their discussion will be taken up later.

The 3.01 per cent of ammoniacal nitrogen in the soil heated at 10 pounds more than doubled in 10 weeks. The amount then dropped somewhat at the end of the 20 weeks. Considering the nitrate results it is seen that not all of this drop is necessarily due to nitrification. In this connection it is interesting to observe that the increase in nitrates in the first 10 weeks was proportionate to the increase in ammonia. In the next 10 weeks, the nitrates increased somewhat. The amino acid nitrogen diminished somewhat in the first 10 weeks but increased to a considerable extent in the next period. From these results and the resuts for ammonia it would seem that ammonification in the first period proceeded quite rapidly at the expense of the amino acid, while in the second period ammonification decreased considerably, allowing a relatively large accumulation of amino acids. The soluble non-protein nitrogen diminished somewhat during the first 10 weeks and then remained stationary during the next period. This group of compounds, although its nature is largely unknown, represent compounds of relatively simple constitution. In the original communication (13) on the determinations of the soluble non-protein nitrogen, it is shown that such complex substances as egg albumin, edestin and casein do not appear to any extent whatever in this group when the soil to which the substances are added is immediately subjected to the analysis. It would seem that the bacteria would attack the compounds in this group rather than the more insoluble portion, but this contention is not borne out entirely by the figures for the analyses. The decrease in the per cent present caused by the incubation is less than the increase of ammonia and nitrate nitrogen. Then, too, the increase in ammonia and nitrates does not represent the total increase, for no doubt relatively large amounts of these compounds were assimilated by the bacteria. The changes taking place in the soils heated to 200° follow in a general way the similar changes in the soil heated in the autoclave at 10 pounds pressure. It is particularly interesting to note the small accumulation of nitrates in

spite of the large amount of ammonia present. Whether this is to be ascribed to actual absence of nitrification or whether the nitrates were being utilized by the bacteria as produced, it is difficult to state. The change in non-protein nitrogen caused by the incubation is only slight.

In table 2 the results for Miami Silt Loam Soil II are found.

From the figures for the loss on ignition it is seen that the soil heated to 200° lost 12.1 per cent of its organic matter. The changes caused by the other treatments are inappreciable. A temperature of 200° also caused a loss of 8.7 per cent of the nitrogen. Fifteen pounds pressure caused a loss of 8.1 per cent of nitrogen while 10 pounds caused a loss of 4.3 per cent of

TABLE 2

Miami silt loam soil II

TREATMENT	LOSS ON IGNITION	TOTAL NITROGEN	PER CENT OF TOTAL NITROGEN							
			Ammonia		Amino acid		Nitrate		Soluble non-protein nitrogen	
			Original basis	Final basis	Original basis	Final basis	Original basis	Final basis	Original basis	Final basis
	per cent	*per cent*								
Air-dry	4.78	0.161	0.89		0.48		0.68		17.1	
100°C.; 2 hours	4.84		1.26		0.57		0.60		21.6	
10 pounds; 9 hours	4.93	0.154	3.16	3.31	2.00	2.13	1.33	1.39	18.3	19.2
15 pounds; 3 hours	4.67	0.148	2.05	2.23	0.92	1.06	0.91	0.99	20.3	22.1
200°C.; 2 hours	4.20	0.147	9.99	10.90	1.22	1.34	0.28	0.31	24.7	27.1
10 pounds; 9 hours; 10 weeks incubation			5.70	5.96	0.45	0.47	1.82	1.90	22.2	23.2
10 pounds; 9 hours; 20 weeks incubation			4.27	4.47	1.25	1.31	2.70	2.82	22.6	23.6
200°C.; 2 hours; 10 weeks incubation			17.10	18.70	0.68	0.74	1.20	1.31	35.8	39.2
200°C.; 2 hours; 20 weeks incubation			20.40	22.50	0.95	1.04	0.97	1.06	34.8	38.1

nitrogen. It is difficult to say from what compounds this loss comes. Nitric acid as such would volatilize wholly at 200° and considerably at the 10-pound and 15-pound treatments. So far as we are able to find, the metallic nitrates are not decomposed at 200°. The loss of nitrates at 200°, therefore, is either to be accounted for by the assumption that at that temperature practically all of the metallic nitrates react with acids to give nitric acid which is volatilized or that the organic matter reduces the nitrates, probably to ammonia. Either assumption seems to be justified from the analytical data and from theoretical considerations. However, this would account for only a small part of the loss of total nitrogen. The chief loss of nitrogen probably comes from the volatilization of ammonia compounds, which in turn are formed by

the degradation of proteins, proteoses, peptones, etc. The purine bases and derivatives no doubt also contribute to the formation of ammonia. The change in the amounts of the different nitrogen compounds caused by the various heat treatments will be taken up later.

The change in ammonia, nitrate, and amino acid nitrogen in the soil treated at 10 pounds pressure caused by the incubation follows, in a general way, the same course as the respective changes in the soil previously discussed. It is interesting to note that the soil incubated 20 weeks contained more amino nitrogen than ammonia. So far as we can recall this is the only soil upon which analyses have been made which shows such a reversal of the usual condition.

TABLE 3

Miami silt loam soil I

TREATMENT	LOSS ON IGNITION	TOTAL NITROGEN	PER CENT OF TOTAL NITROGEN							
			Ammonia		Amino acid		Nitrate		Soluble non-protein nitrogen	
			Original basis	Final basis	Original basis	Final basis	Original basis	Final basis	Original basis	Final basis
	per cent	*per cent*								
Air-dry	3.75	0.130	1.04		0.36		1.43		17.95	
100°C.; 2 hours	3.54		1.16		0.52		1.41		17.7	
10 pounds; 9 hours	3.65	0.127	2.05	2.11	0.72	0.74	2.52	2.59	21.2	21.8
15 pounds; 3 hours	3.80	0.126	1.95	2.01	0.61	0.63	2.22	2.28	19.5	20.2
200°C; 2 hours	2.90	0.117	4.61	5.14	0.53	0.60	0.41	0.46	32.0	36.4
10 pounds; 9 hours; 10 weeks incubation			3.62	3.70	0.51	0.52	6.4	6.6	20.5	21.0
10 pounds; 9 hours; 20 weeks incubation			1.74	1.78	1.92	1.96	6.2	6.3	22.7	23.3
200°C.; 2 hours; 10 weeks incubation			12.0	13.30	0.63	0.70	2.02	2.25	38.15	42.4
200°C.; 2 hours; 20 weeks incubation			9.6	1.06	0.94	0.93	4.78	5.31	27.9	31.0

The changes in the incubated 200°-heated soils follow in some respects the same course as the respective changes in the Calhoun silt loam. The most noteworthy difference is to be found in the non-protein nitrogen which nearly doubles in the first 10 weeks but drops somewhat in the next period. Part of the first increase is due, of course, to the increased ammonia which would be in this fraction.

In table 3 are given the results obtained with the second sample of Miami silt loam soil. With this soil the loss of organic matter of the 200°-heated soil as shown by the loss on ignition was 22.6 per cent, as compared with a loss of 12.1 per cent for the other sample of this soil type. The difference is all the more noteworthy when it is recalled that the one showing the less loss

of organic matter contains at the start about 1 per cent more total organic matter. The loss of nitrogen of the soil the results of which are given in table 3 is 10.0 per cent.

With the exception of the nitrate nitrogen the changes which the various forms of nitrogen exhibit in the soil heated at 10 pounds and inoculated and incubated are in general similar to the changes undergone by the other sample of Miami soil. The nitrates show a gradual rise at the end of the first and second 10 weeks when compared with the amounts present before incubation. In the case of the 200°-heated soils the changes are not consistent when compared with the similar changes of the soils previously discussed. The amounts

TABLE 4

Peat soil

TREATMENT	LOSS ON IGNITION	TOTAL NITROGEN	PER CENT OF TOTAL NITROGEN							
			Ammonia		Amino acid		Nitrate		Soluble non-protein nitrogen	
			Original basis	Final basis	Original basis	Final basis	Original basis	Final basis	Original basis	Final basis
	per cent	*per cent*								
Air-dry	47.3	3.39	0.23		0.260		0.37		4.96	
100°C.; 2 hours	48.3		0.25		0.190		0.28		7.7	
10 pounds; 9 hours	47.2	3.09	1.45	1.59	0.220	0.24	0.43	0.47	6.43	7.05
15 pounds; 3 hours	47.0	2.39	5.95	8.43	0.190	0.27	0.31	0.44	6.53	9.25
200°C.; 2 hours	41.6	2.42	1.35	1.90	0.170	0.24	0.05	0.07	10.90	15.30
10 pounds; 9 hours; 10 weeks incubation			1.28	1.40	0.135	1.48	3.92	4.30	6.66	7.31
10 pounds 9 hours; 20 weeks incubation			1.45	1.59	0.140	0.15	4.17	4.58	4.33	4.75
200°C.; 2 hours; 10 weeks incubation			1.02	1.42	0.110	0.15	3.76	5.26	5.78	8.10
200°C.; 2 hours; 20 weeks incubation			1.55	2.17	0.190	0.27	2.00	2.80	5.43	7.60

of ammonia and non-protein nitrogen have been increased by the incubation, whereas the amino acid nitrogen was diminished. The nitrates were increased by both periods of incubation but the increase was less for the longer period than for the shorter.

The results for the peat soil appear in table 4. It is observed that the treatment at 200° caused a loss of 12.05 per cent of organic matter, whereas by the same treatment 28.6 per cent of nitrogen was lost. This is a far greater loss of nitrogen than the three mineral soils have undergone. On first thought it would seem that there should be less loss of nitrogen from a peat soil, the nitrogen of which is generally held to be in a relatively inert, unavailable form. The reason for the greater loss probably lies in the fact that in the

peat soil there is a less proportionate amount of the minerals which act as adsorbents or absorbents for ammonia. For, as stated above, it probably is largely as ammonia that the nitrogen is lost. It is seen that in the peat soil the amount of ammonia found in the 200°-heated soils is low when compared with the similar amounts in the mineral soils.

When the results for the incubated soils are examined it is seen that but little change was brought about by the incubation. The results for the peat soil are striking in that respect when compared with the mineral soils. Probably the explanation of the small amount of change brought about by incubation is that the heat treatment released soluble toxic material to a great extent in this soil, so high in organic matter. Another interesting point seen on an inspection of the results for the peat soil is that in many cases the sum of the nitrate, ammonia and amino acid nitrogen is nearly equal to the soluble non-protein nitrogen. Certainly all the nitrate and amino acid nitrogen would be in the soluble, non-protein nitrogen fraction and the larger portion, if not all of the ammonia, would be in this fraction also. Therefore, unlike the other soils, the soluble non-protein nitrogen includes but little material other than ammonia, nitrate and amino acid nitrogen.

The changes in the various forms of nitrogen which the different heat treatments brought about have been plotted and appear in figures 1 to 4. As is seen from an inspection of the figures, treatments are plotted on the ordinates and the per cent change on the abscissa. Different standards for the different constituents have been used for the latter. Because the treatment points are separated from each other by regular periods on the ordinate, it is not meant to convey that actually, for instance, there is the same difference between air-dry soil and soil heated for 2 hours at 100°C., as there is between soil given the latter treatment and heated in the autoclave for 9 hours at 10 pounds pressure.

The results for the ammonia in the different soils are shown in figure 1. Heating at 100° for 2 hours is seen to increase slightly the amount of ammonia in all the soils and 9 hours at 10 pounds in the autoclave increases the ammonia still more. Fifteen pounds for 3 hours does not give such an increase in the mineral soils as the 10-pound treatment. Evidently the length of time of heating has quite an appreciable effect. We do not have an explanation of the great increase in ammonia in the peat soil. At 200° all the soils show an increase in ammonia when compared with the treatment at 15 pounds, except the peaty soil which shows a decrease. This decrease, as pointed out above, is probably largely due to the fact that the ammonia had volatilized from the peat soil to a considerable extent. While ammonia has no doubt been evolved from the mineral soils to a considerable extent at 200° also, yet the large amount of minerals in the latter soils makes necessary a greater accumulation of ammonia before appreciable volatilization takes place.

The amino acid results are shown in figure 2. The amino acid nitrogen in the two Miami silt loam soils was increased by heat at 100°, whereas

this form of nitrogen was decreased in the peat and Calhoun soils by the same treatment. Treatment at 10 pounds increased the amount of amino acid nitrogen in all the soils and at 15 pounds the amounts were decreased.

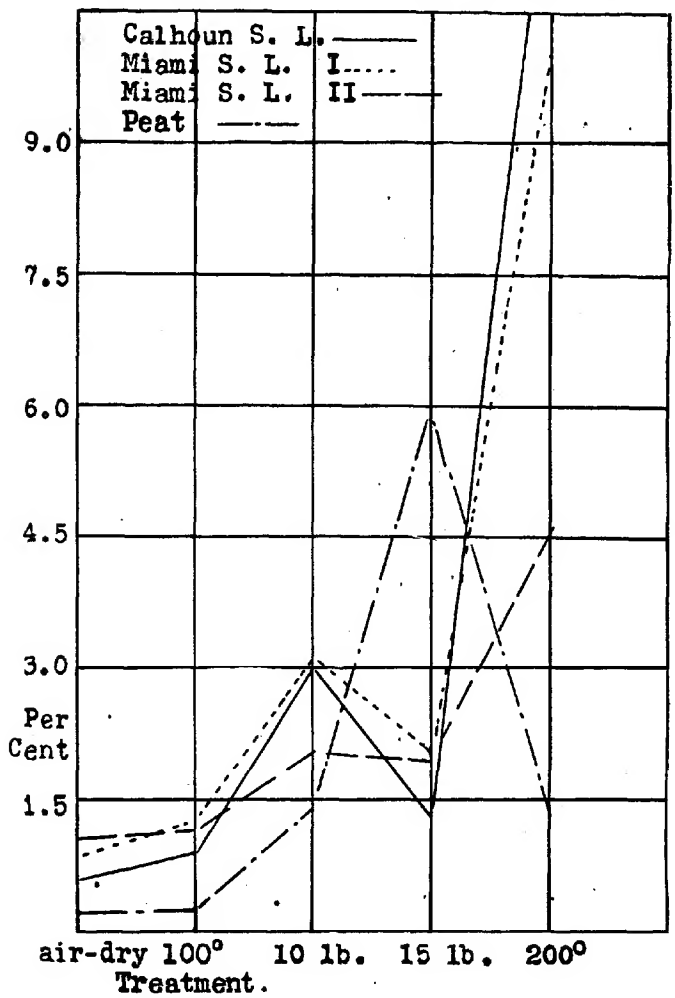

FIG. 1. DIAGRAM SHOWING EFFECT OF HEAT ON AMMONIA IN THE SOILS STUDIED

At 200° the Calhoun soil and the Miami soil I, showed an increase and the other soils a decrease. The two soils that had an increase in amino acid nitrogen also showed the greatest increase in ammonia. Another interesting point is the slight change in amino acid nitrogen shown by the peat soil.

Figure 3 shows the results for the nitrate nitrogen. Perhaps the most interesting thing shown by the figure is that the curves for the various soils are arranged in the order of the per cent of total organic matter present in the

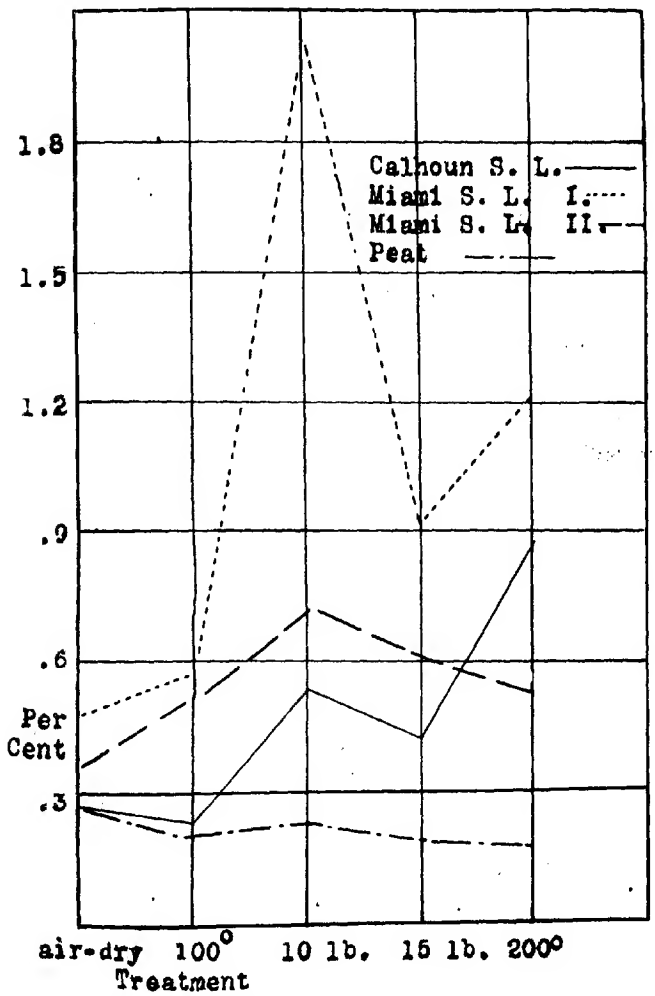

FIG. 2. DIAGRAM SHOWING EFFECT OF HEAT ON AMMONIA ACIDS IN THE SOILS STUDIED

soils. The peat soil, containing the highest amount, being at the bottom and the Miami silt loam soil, containing the least, at the top. Heat at 100° caused a decrease in nitrates in all the soils but the Calhoun silt loam. Ten pounds and 15 pounds in the autoclave caused an increase in the amounts of

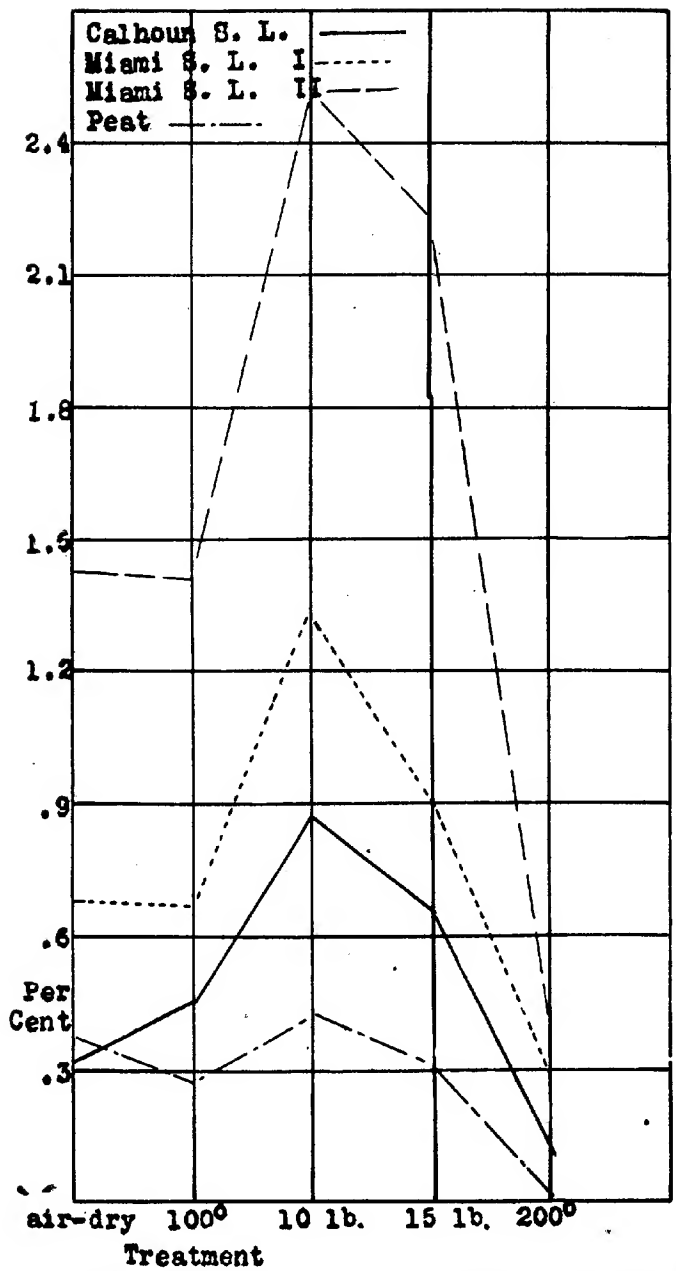

FIG. 3. DIAGRAM SHOWING EFFECT OF HEAT ON NITRATE NITROGEN IN THE SOILS STUDIED

nitrates in all the soils, the 10 pounds a greater increase than the 15 pounds. This is contrary to the findings of Lyon and Bizzell (7, 8) who found that nitrates were greatly reduced by heat at 15 pounds for 2 hours. At 200° for 2 hours it is seen from the figure that the nitrates have been greatly reduced. As pointed out earlier in this paper, this decrease is due to volatilization of nitric acid or reduction of the nitrates by the organic matter.

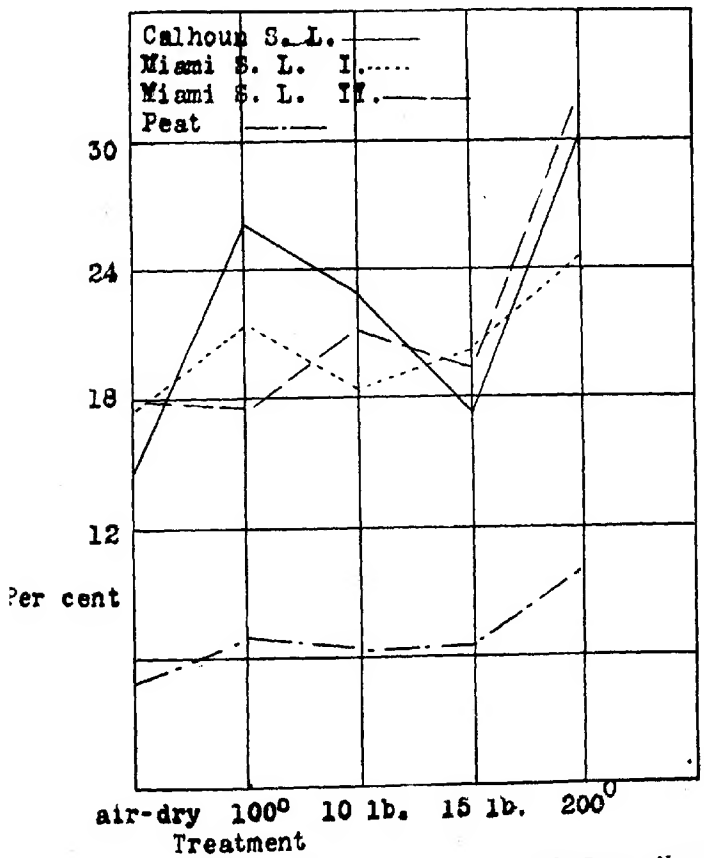

FIG. 4. DIAGRAM SHOWING EFFECT OF HEAT ON SOLUBLE NON-PROTEIN NITROGEN IN THE SOILS STUDIED

The soluble non-protein nitrogen results are shown in figure 4. It is seen that the curves do not resemble very closely either the curves for the ammonia or the nitrate results. This is important in that it shows that when we follow the changes of nitrate and ammonia in a soil we have only half-truths, for while these two substances are perhaps the most important nitrogenous constituents of the soil in that they are used more than any other

compounds as plant nutrients, yet our results show that there is a rather large class of soluble nitrogenous compounds other than the nitrates and ammonia whose transformation does not follow the latter two compounds. In this connection, the results for the soluble non-protein nitrogen for both the incubated and unincubated peat soils are particularly interesting. It is observed that in some cases the sum of the nitrate, amino acid and ammonia nitrogen is nearly equal to, and in one case exceeds, the total soluble non-protein nitrogen. In this one case noted no doubt there must be some error of analysis involved. One source of error in the determination of the soluble non-protein nitrogen is a possibility of the loss of ammonia from the 1½ per cent sodium hydroxide used for the extractive agent. This would be particularly true for these treated soils which contain such large amounts of ammonia.

SUMMARY AND CONCLUSIONS

The following conclusions may be drawn from the soils receiving simply the respective heat treatments.

1. The amount of ammonia was increased by all the heat treatments, the higher temperatures to which the soils were heated giving in general greater increases. At 10 pounds the increase was greater than at 15 pounds, the reason being the longer time the soils were heated at 10 pounds.

The peat soil heated to 200°C. contained less ammonia than the soil heated at any other temperature, the reason being the excessive volatilization of ammonia at that temperature.

2. The amino acid nitrogen results in general followed those for ammonia nitrogen. The peat soil again is an exception, not much of any change being brought about by any of the treatments.

3. Not much change was brought about in the nitrate content by dry heat at 100°C., 10 pounds in the autoclave for 9 hours increased the amount considerably in every soil. Fifteen pounds for 3 hours caused a somewhat less increase than the 10-pound treatment. A temperature of 200°C. caused almost the total disappearance of all nitrates.

4. All the heat treatments caused an increase in soluble non-protein nitrogen. The least change was brought about in the peat soil. This is probably in part due to the fact that considerable amounts of ammonia escaped from the soil at the higher temperature treatments.

The following conclusions may be drawn from the results for the soils heated to 10 pounds in the autoclave for 9 hours and the soil heated to 200°C. for 2 hours, both later inoculated and incubated as stated in the first part of this paper.

1. In the three mineral soils heated to 10 pounds for 9 hours and with 10 weeks' incubation after inoculation with fresh soil, there was an increase of ammonia. Twenty weeks' incubation likewise caused an increase in two

cases, though not such an increase as the 10-week period. Twenty weeks' incubation of the Calhoun soil caused a slight decrease in ammonia. The amount of ammonia in the peat soil after the two different heat treatments was not greatly influenced by either incubation periods.

2. The changes in the amino acid content of the soils after incubation were in general as follows. Ten weeks' incubation caused a decrease in amino nitrogen. There was always more amino nitrogen at the end of 20 weeks than at the end of ten. In two of the soils there was more amino nitrogen at the end of the 20 weeks than there was at the beginning.

3. Nitrates were invariably increased by the two incubation periods, the longer period usually giving the greater increase.

4. The amount of non-protein nitrogen was not changed very much by incubation of the mineral soils which had been heated to 10 pounds. In the peat soil 20 weeks decidedly lessened the amount of this group of compounds. In the mineral soils heated to 200°C. there was always an increase in soluble non-protein nitrogen after incubation. Here also the peat soil showed a decrease for both incubation periods.

REFERENCES

(1) Burgess, P. S. 1913 The aluminum reduction method as applied to the determination of nitrates in "Alkali" soils. *In* Univ. Cal. Pub. Agr. Sci., v. 1, pp. 51–62.

(2) Deherain, P. P., and DeMoussy, M. E. 1896 Sur l'oxydation de la matière organique du sol. *In* Ann. Agron., t. 22, p. 306–337.

(3) Greenwald, I. 1915 The estimation of non-protein nitrogen in blood. *In* Jour. Biol. Chem., v. 21, p. 61–63.

(4) Kelley, W. P., and McGeorge, M. 1913 The effect of heat on Hawaiian soils. *In* Hawaii Agr. Exp. Sta. Bul. 30, p. 37.

(5) Kober, P. A., and Sugiura, K. 1915 A micro-chemical method for the determination of alpha and beta amino acids and certain derivatives. *In* Jour. Amer. Chem. Soc., v. 37, p. 1547–1584.

(6) Kopeloff, N., and Coleman, D. A. 1917 A review of investigations in soil protozoa and soil sterilization. *In* Soil Sci., v. 3, p. 197–271.

(8) Lyon, T. L., and Bizzell, J. A. 1913 Water-soluble material in soils sterilized and reinoculated. N. Y. Agr. Exp. Sta. (Cornell) Bul. 326.

(7) Lyon, T. L., and Bizzell, J. A. 1910 Effect of steam sterilization on the water-soluble matter in soils. N. Y. Agr. Exp. Sta. (Cornell) Bul. 275, p. 130–155.

(9) Pickering, S. U. 1908 The action of heat and antiseptics on plant growth. *In* Jour. Agr. Sci., v. 3, p. 32–54.

(10) Pickering, S. U. 1908 Studies on germination and plant growth. *In* Jour. Agr. Sci., v. 2, p. 411–434.

(11) Potter, R. S., and Snyder, R. S. 1915 The determination of ammonia in soils. *In* Jour. Indus. Engin. Chem., v. 7, p. 221–226.

(12) Potter, R. S., and Snyder, R. S. 1915 The amino acid nitrogen of soil. *In* Jour. Indus. Engin. Chem., v. 7, p. 1049–1055.

(13) Potter, R. S., and Snyder, R. S. 1916 The soluble non-protein nitrogen of soil. *In* Jour. Agr. Research, v. 6, p. 61–64.

(14) Richter, L. 1896 Uber die Verandernungen welche der Boden durch das Sterilisieren erleidet. *In* Landw. Vers. Sta., Bd. 47, p. 269–274.

(15) RUSSEL, E. J., AND HUTCHINSON, H. B. 1909 The effect of partial sterilization of soil on the production of plant food. *In* Jour. Agr. Sci., v. 3, p. 111–144.

(16) RUSSEL, E. J., AND HUTCHINSON, H. B. 1913 The effect of partial sterilization of soil on the production of plant food. *In* Jour. Agr. Sci., v. 5, p. 152–221.

(17) SCHREINER, O., AND LATHROP, E. C. 1912 The chemistry of steam-heated soils, U. S. Dept. of Agr. Bur. Soils Bul. 89.

DETERMINATION OF LIME AND PHOSPHORIC ACID IN PEAT SOILS: COMPARISON OF JÖNKÖPING WITH BREMEN METHOD[1]

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INTRODUCTION

In this country the analysis of peat soils has, up to the present time received little attention, as is well illustrated by the failure of the Association of Official Agricultural Chemists to recognize the need of any special methods for the analysis of these as distinct from the mineral or ordinary soils (1, p. 17–29). The Germans have long since recognized such a difference, for the former (4, p. 111–128) prescribing methods quite distinct from those employed with the latter (4, p. 1–110).

During the past 30 to 40 years, a great number of peats have been analyzed in Germany and Sweden, the number in the latter country reaching, according to von Feilitzen (3, p. 164), a total of 3700 samples as early as 1903. This work was begun at the Bremen station and the special method developed there was followed in the Jönköping laboratory of the Swedish Peat Land Cultivation Society for the first few years after it was established in 1886. But another method for the determination of the mineral constitutents of these soils was soon developed and this has since been used exclusively there. We shall refer to this as the Jönköping method.

The only previous comparison of the two methods reported is one from the Jönköping laboratory in 1892 (2, p. 452–457). As a large number of peat soils are being analyzed in this laboratory it appeared desirable to make a further comparison, especially as a recent study by one of us (6) caused us to suspect that the iron content might have an important influence upon the amount of phosphoric acid recoverable by the Jönköping method.

Bremen method

According to the Bremen method (4, p. 118–119), used in general by the German peat experimental stations, a sample of peat is incinerated and the ash extracted with aqua regia. In a hydrochloric acid solution of the material rendered soluble in this manner the percentages of lime and phosphoric acid are determined by the usual procedure.

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Jönköping method

This method has been employed by the Swedish Peat Land Cultivation Society for the analysis of all peat soils since 1890 (3, p. 164). It consists (5) in digesting at room temperature a quantity of air-dried soil equivalent to about 70 gm. of water-free material with 700 cc. of 12 per cent hydrochloric acid for 48 hours. At the end of this time the extract is filtered twice, first through linen and then through filter paper and from the solution thus obtained 50-cc. portions (5 gm.) are taken for the determination of lime and phosphoric acid as well as the oxides of iron and aluminum.

Lime (5, p. 22). A 50-cc. portion of the acid extract is evaporated to dryness and the residue ignited over a Bunsen burner, at first gently, more strongly toward the end, but avoiding any intense ignition. As soon as the contents of the dish have started to glow it is covered with a thin asbestos disc and the ignition continued for three-quarters of an hour, after which the crust formed is broken up with a glass rod and the ignition continued half an hour longer, or until no carbon is to be observed on the sides of the dish or in the ignited mass. As soon as it is free of carbon the residue is pulverized by means of a heavy glass rod or agate pestle, treated with water and a pinch of sal ammoniac, and the mixture digested on the water bath for an hour with occasional stirring. At the end of this time the liquid is filtered off, the residue extracted twice with boiling water, and finally transferred to a filter and washed. The lime is determined in the filtrate, using the permanganate method.

Phosphoric acid (5, p. 26). Another 50-cc. portion of the acid extract is evaporated and incinerated as described above, the residue extracted with hydrochloric acid, filtered, evaporated to dryness and finally taken up with 25 per cent nitric acid. The phosphorus in this solution is determined volumetrically, a normal solution of sodium hydroxide being used as a solvent for the yellow precipitate, and the excess titrated with normal nitric acid, with the factor 1 cc. of normal sodium hydroxide = 0.00315 gm. P_2O_5.

DATA OBTAINED BY C. VON FEILITZEN

There appears to be only one publication that deals with the relative amounts of the various constituents obtained by the above-mentioned methods. Von Feilitzen (2, p. 449–457) in 1892 reported a comparison in the case of 11 Swedish peats and mucks. The portions of his data bearing on the present subject have been assembled in table 1.

It is to be observed that the amount of lime obtained by the two methods is, for practical purposes, about the same, it being slightly the lower with the Jönköping method, but that only from one-half to less than one-third as much phosphoric acid is obtained by the latter method. While the amounts of the oxides of iron and aluminum do not differ widely, if the three muck

soils, the last in the list, are left out of the comparison, the Bremen method gives the larger amount in every case.

EXPERIMENTAL

Eight samples of peat soil, varying in lime content from 0.6 per cent to 3.9 per cent, were used in this study. The composition, depth and source of each sample are shown in table 2.

TABLE 1

Comparison of the Bremen (B) and Jönköping (J) methods for the analysis of peat and muck soils, as shown by data obtained by von Feilitzen

SAMPLE	DEPTH	VOLATILE MATTER	Fe_2O_3	Al_2O_3	CaO		P_2O_5	
			B	J	B	J	B	J
	cm.	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
Highmoor, Flahult	0–30	98.05	0.24	0.16	0.21	0.16	0.11	0.05
	30–60	98.60	0.29	0.22	0.19	0.21	0.06	0.03
	60–90	98.59	0.30	0.30	0.17	0.18	0.06	0.02
	90–120	98.58	0.46	0.39	0.13	0.13	0.07	0.02
	120–150	98.63	0.71	0.57	0.11	0.10	0.07	0.01
Lowmoors, Flahult	0– 20	88.85	3.09	2.74	0.04	0.08	0.15	0.07
Martebo myr	0– 20	83.38	2.00	1.40	8.48	8.64	0.12	0.04
Vårgårda	0– 20	70.71	1.45	0.78	1.35	1.17	0.10	0.03
Vargsäter	0– 20	46.32	10.12	5.12	2.89	2.71	0.25	0.10
Skeppsholmen	0– 20	25.59	7.39	4.57	0.61	0.28	0.30	0.13
Högs gård	0– 20	21.02	8.52	3.92	0.59	0.22	0.22	0.10

TABLE 2

Comparison of the Bremen (B) and the Jönköping (J) methods in the analysis of Minnesota peat soils

SAMPLE	SOURCE OF SAMPLE, COUNTY	DEPTH	VOLATILE MATTER	$Fe_2O_3+Al_2O_3$ J	CaO		P_2O_5	
					B	J	B	J
		inches	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
1	Anoka	0–8	74.3	5.66	3.91	3.35	0.55	0.34
2	Anoka	0–8	77.8	4.21	3.01	2.60	0.42	0.20
3	Anoka	0–8	77.4	3.20	1.36	1.10	0.30	0.10
4	Anoka	0–8	83.4	3.95	2.25	1.94	0.33	0.10
5	Ramsey	0–8	83.8	1.22	1.24	0.94	0.32	0.11
6	Itasca	9–16	90.7	1.55	0.84	0.60	0.20	0.03
7	Blue Earth	9–16	78.0	1.36	2.79	2.50	0.23	0.07
8	Itasca	0–8	89.5	1.10	0.80	0.55	0.25	0.09

In order to be able to compute the amount of the moist peat equivalent to 70 gm. of dry material, as specified in the Jönköping method, duplicate moisture determinations were made by drying small samples for 24 hours in a water oven. In adding the 12 per cent hydrochloric acid the water in the peat was taken into consideration and allowance made for this, enough con-

centrated hydrochloric acid being added after the bulk of the 12 per cent acid had been put on the sample to bring the final strength of the hydrochloric acid up to the required 12 per cent. The digestion was carried out in liter bottles with frequent shaking. At the end of the 48-hour period the supernatant liquid was filtered off and the analyses made as described above.

In the Bremen method 5-gm. samples were weighed out in silica dishes and ignited, at first very gently but finally in the muffle. The ash thus obtained was evaporated with 25 cc. of aqua regia to complete dryness twice and then taken up with hydrochloric acid, diluted to 500 cc. with water and an aliquot part of this taken for each determination.

The eight samples were analyzed by both of the above-described methods. The results are shown in table 2. The same general observations made in regard to the work reported by von Feilitzen (table 1) may be applied to these. While the amount of lime obtained by the Bremen method is in every case greater than that obtained by the Jönköping, the difference in amount is not so great but that the percentage of this constituent found adequate for plant growth by the former should also prove adequate when the determination is made by the latter.

The phosphoric acid obtained by the Bremen method is from two to three times greater than that obtained by the Jönköping. Von Feilitzen (3, p. 160–162) has already pointed out that the difference in the amount of this constituent obtained by the two methods constituted their chief difference, claiming that the smaller amount obtained more truly represented that actually available for plant growth since much of the phosphoric acid found when a peat is ashed is not available for plant use.

Completeness of recovery of lime and phosphoric acid

The completeness of recovery of the lime and phosphoric acid in the acid extract obtained by the Jönköping method was tested. The data are reported in table 3. It is to be seen that not the whole of the lime is recovered, in most cases sufficient being found in the residue to make the total amount extracted by the acid equal to that found by the Bremen method. As a measure of precaution we had extracted the residue five times with hot water instead of only twice, as is done in the Jönköping laboratory. So the shortage cannot be attributed to insufficient washing of the residue preceding its removal from the dish to the filter. It is of interest to note that the amount found in the residue varies directly with the percentage of oxide of iron. In general the higher the percentage of this constituent the larger was the amount of lime found in the residue. There is no definite relation between the amount of lime in the residue and the amount of oxide of aluminum.

Practically the whole of the phosphoric acid extracted by the 12 per cent hydrochloric acid is recovered, only a negligible quantity for the most part

being left in the residue. It will be seen that in the cases where the larger amounts are found in the residue the sample carries a comparatively high percentage of iron oxide. One of us has already shown (6, p. 306–307) that phosphoric acid may be held in the residue when this has been heated too strongly, as a result of the formation of difficulty-soluble phosphates of iron and aluminum. There appears to be no definite relation between the amount of oxide of iron present and the amount of phosphoric acid extracted.

SUMMARY

1. Two methods for the determination of lime and phosphoric acid in peat soils are compared, viz., the Bremen, in which the sample is incinerated and the ash extracted with aqua regia, and the Jönköping, in which an air-dry sample is extracted with 12 per cent hydrochloric acid.

TABLE 3

Analysis of Minnesota peat soils by the Jönköping method, showing the completeness of recovery of lime and phosphoric acid from the acid extract

SAMPLE	Al_2O_3	Fe_2O_3	LIME (CaO)			PHOSPHORIC ACID (P_2O_5)		
			Original extract	Residue from extract	Total amount extracted	Original extract	Residue from extract	Total amount extracted
	per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
1	1.89	3.77	3.35	0.67	4.02	0.34	0.020	0.360
2	1.69	2.52	2.60	0.37	2.96	0.20	0.014	0.214
3	1.35	1.85	1.10	0.28	1.38	0.10	0.004	0.104
4	2.38	1.57	1.94	0.21	2.15	0.10	0.003	0.103
5	0.79	0.43	0.94	0.18	1.12	0.11	0.002	0.112
6	1.18	0.37	0.60	0.05	0.65	0.03	0.003	0.033
7	1.03	0.33	2.50	0.13	2.63	0.07	0.005	0.075
8	0.80	0.30	0.55	0.14	0.69	0.09	0.002	0.092

2. With the former, larger amounts of iron and aluminum oxides, lime and phosphoric acid are obtained, the difference with the first three constituents being small, but that of the last very large.

3. The smaller amount of lime found by the Jönköping method is due to part of that extracted being rendered insoluble in water in the course of the analysis and hence not included in the final precipitation. This unrecovered portion varies directly with the amount of iron present. Practically all of the phosphoric acid is recovered from the acid extract.

REFERENCES

(1) Association of Official Agricultural Chemists 1916 Methods of analysis. *In* Jour. Assoc. Off. Agri. Chem., v. 1, no. 4, pt. 2, p. 17–29.

(2) VON FEILITZEN, CARL 1892 Forvaltningstutskottets berättelse till revisorerna öfver räkenskaper och förwaltning 1891. *In* Svenska Mosskulturför. Tidskr., v. 6, no. 5, p. 442–481.

(3) VON FEILITZEN, HJALMER 1910 Über die chemische analyse des Moorbodens bei der Bewertung für Kulturzwecke. *In* Verhandlungen der Zweiten Internationalen Agrogeologenkonferenz, p. 160–168. Stockholm.

(4) KÖNIG, J. 1911 Untersuchung Landwirtschaftlich und Gewerblich Wichtiger Stoffe. Ed. 4, 1226 p. 426 fig. Berlin.

(5) LUGNER, IVAR 1912 Metod för undersökning af torfjord för kulturändamal, anväud vid Svenska Mosskulturföreningens kemiska laboratorium. *In* Berättelse öfver Verksamheten vid Svenska Mosskulturföreningens Kemiska Laboratorium år 1911, p. 22–26.

(6) ROST, CLAYTON O. 1917 The determination of soil phosphorus. *In* Soil Sci., v. 4, p. 295–311.

POTASSIUM REQUIREMENTS OF BACTERIA[1]

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INTRODUCTION

The knowledge that plants require certain mineral constituents in order to continue their life processes, dates back to the time of Liebig (6). It was, however, not definitely known until 1870 just what these essential salts are. In that year Nobbe (7) and his associates, through careful investigation, demonstrated to what extent plant activities would continue in the absence of the various elements. The proof that plants of the lower order require the same essential mineral constituents is now quite well established.

The literature relating to the influence of certain inorganic salts on biological activities is voluminous. Some of the researches on the subject have been referred to in another paper by the writer (2). In the data set forth by Gerlach and Vogel (1) in 1903, and by Vogel (10) in 1912, these investigators demonstrated the extent to which microbial activities (fixation of nitrogen by *Azotobacter chroococcum*) take place when certain of the essential elements are not supplied. In a similar way Krzemieniewska (5) in 1910 showed that K, Ca, Mg, P and S were all essential to the development of *Azotobacter*. In these experiments the physical factor, namely, the effect upon the microbial activities in varying the total osmotic concentration of the various solutions when demonstrating the effect of the various cations and anions, was not considered. Also, the antagonism of the various salts was not observed.

Since it has been demonstrated by the respective methods of the above investigators that potassium is one of the essential inorganic cations for bacteria (nitrogen-fixing) it was deemed of scientific importance to verify these results, if possible, by methods in which the physical factors were controlled. Still further, it appeared desirable to ascertain to what extent potassium is a factor. In other words, how much of this element do microorganisms require in order to carry on their life processes?

EFFECT OF VARIATION IN TOTAL CONCENTRATION AND LACK OF POTASSIUM UPON BIOLOGICAL ACTIVITIES

Tottingham (9) and Shive (8), working with plants, and the writer (3), in a previous work with microorganisms, demonstrated to what extent the

[1] Paper read at the meeting of the Society of American Bacteriologists, at Washington, D. C., December 27, 1917.

total concentration (all other factors, as the arrangement of salts, etc., being constant) influences life processes. At a very low concentration (total 0.10 atmosphere) the writer found practically no difference in the formation of ammonia from dialyzed peptone by *Bacillus subtilis* in the various combinations of the three salts. For this reason, and also since a considerable amount of ammonia was formed in all the solutions of the sub-optimal series (total concentration 0.10 atmosphere) and the amounts corresponded very favorably with the amounts formed in the optimal series (total concentration 1.0 atmosphere), this concentration (0.10 atmosphere) was adopted as the most desirable for investigations to ascertain the requirements of microorganisms.

At such a low concentration as 0.10 atmosphere of diffusion tension, keeping the salts in their same proportion, to what extent is it possible to vary the total concentration of the solutions without altering the results? An experiment was performed to ascertain if it is possible to reduce the total concentration from 0.1 to 0.09 atmosphere without materially affecting the ammonia production.

The Gibbs triangle scheme which was successfully employed in previous biological work (3, 4) was used. In this case, however, it was only to make use of the solutions appearing on the left-hand side of the triangle where the potassium sulfate furnishes but one-tenth of the total diffusion tension, and the other two salts are in various proportions. These solutions are indicated as R_1C_1, R_3C_1, R_5C_1, R_7C_1 and R_8C_1.

Three series of each of the solutions, R_1C_1, R_3C_1, etc., were prepared. In series I, the solutions were identical to the corresponding solutions of the sub-optimal series of the previous work (3), in which the total concentration of the three salts added was 0.10 atmosphere. In series II the three salts were all in the same proportion as the corresponding solutions in series I, but the total amount of salt was one-tenth less than in series I. In series III the proportion of the two salts magnesium sulfate and calcium phosphate was the same as in the other two series, the total amount of each the same as in series I, and the total osmotic concentration the same in series II. To test the biological activity in the absence of potash, in the third series potassium sulfate was left out of the combination.

The method of calculating and preparing solutions of known concentration was the same as is given in detail in the previous paper (3). Similarly dialyzed peptone, the equivalent of 100 mgm. of nitrogen, of the same stock solution was used. As before, the solutions were inoculated with a pure culture of *B. subtilis* and incubated for 7 days at 30°C., at the end of which period the ammonia determinations were made. All determinations were made in duplicate or triplicate, the average of which is recorded in table 1. The experimental error was never more than 3.5 per cent.

The results for series I as presented in the table 1 demonstrate that there was the same amount of ammonia formed in the various solutions composed of the different combinations of the three salts. This corroborates the results

previously obtained by the writer. In like manner, the same amount of ammonia was realized in the various solutions of series II. This was also true in the third series. As the results of series I are practically the same as those of series II, we would conclude that reducing the total concentration from 0.10 to 0.09 atmosphere, while keeping the salt proportion the same, under the conditions of the experiment would not cause any difference in the biological activities.

TABLE 1

Effect of different concentrations of various combinations of 3-salt solutions and similar solutions without potassium sulfate upon the formation of ammonia from dialyzed peptone by Bacillus subtilis

SERIES NO.	SOLUTION NO.	SALT PROPORTION	TOTAL CONCENTRATION	AMOUNT OF AMMONIA FORMED
				mgm. N
1	R_1C_1	$1MgSO_4$ $1K_2SO_4$ $8Ca(H_2PO_4)_2.2H_2O2H_2O$	0.10	34.0
1	R_3C_1	$3MgSO_4$ $1K_2SO_4$ $6Ca(H_2PO_4)_2.2H_2O2H_2O$	0.10	33.2
1	R_5C_1	$5MgSO_4$ $1K_2SO_4$ $4Ca(H_2PO_4)_2.2H_2O2H_2O$	0.10	33.5
1	R_7C_1	$7MgSO_4$ $1K_2SO_4$ $2Ca(H_2PO_4)_2.2H_2O2H_2O$	0.10	33.6
1	R_8C_1	$8MgSO_4$ $1K_2SO_4$ $1Ca(H_2PO_4)_2.2H_2O2H_2O$	0.10	33.2
2	Same as in series I....	Same as in series I.	0.09	33.6*
3	Same as in series I....	Same as in series I, except no K_2SO_4	0.09	9.0
Check (peptone alone)				9.0

* As the results of the various determinations of series II were all practically the same, as was also the case in series III, it is not necessary to repeat all the figures.

In comparing the results of series III with those of the other two series, it is apparent that potassium is the limiting factor. While in series I and II we have the same arrangement of salts but a difference of 0.01 atmosphere in total concentration, the same amounts of ammonia were formed in both cases. Hence the slight variation in the partial concentration of each individual salt and the difference in the total concentration of the solution were

not influencing factors. In series I, the partial diffusion tension furnished by magnesium sulfate and calcium phosphate is the same as in series III, and the two series differ only in the total concentration, which is 0.01 atmosphere less in series III and in the absence of potassium sulfate in the latter. That the slight difference in total concentration does not influence the results is apparent from a comparison of series I and II, hence the difference of 24.5 mgm. of nitrogen produced in the two series must be attributed to the lack of potassium.

Also, the addition of magnesium sulfate and calcium phosphate, with a concentration of 0.09 atmosphere, without applying potassium sulfate, did not increase the ammonia formation over that of the check (peptone alone). A chemical analysis of the dialyzed peptone which was used showed the presence of the following: 0.02 per cent P_2O_5; 0.0044 per cent SO_3, 0.0024 per cent K_2O, 0.0067 per cent CaO, and 0.00234 per cent MgO. Since 5 cc. of the peptone furnishing 100 mgm. of nitrogen was used, 0.1 mgm. of K_2O was added in each determination. As is shown in series III, this amount of potash furnished sufficient potassium to allow the formation of 9.0 mgm. of ammonia.

That the cation K and not the anion SO_3 was the limiting factor, since there was sufficient SO_3 furnished by magnesium sulfate in series III to supply the needs of the organisms, has already been demonstrated in previous experiments. In the sub-optimal series, practically the same amounts of ammonia were formed irrespective of the amounts of the various salts and SO_3.

THE EXTENT OF POTASSIUM REQUIREMENTS BY BACTERIA

With the above data at hand, an experiment was performed to ascertain how much potassium organisms require to fulfil their needs. The total concentration of the solution used varied from 0.10 to 0.09 atmosphere. Four sets of three solutions each were prepared as before. In all cases magnesium sulfate furnished one-tenth and calcium phosphate eight-tenths of the total concentration. In the first set potassium sulfate furnished one-tenth of the concentration, in the second set one-half as much potassium sulfate was added as in the first, in the third one-fifth as much, and in the fourth set no potassium sulfate was added. A set of check determinations were made at the same time.

From the data in table 2 it is evident that the organism used (*B. subtilis*) obtained sufficient potassium from 0.24 mgm. of K_2O in 100 cc. of solution to carry on its maximum activities for 7 days at 30°C. Moreover, additions of two and one-half and five times this amount of potash were not effective, within experimental error, in increasing the ammonia formation. The addition of but 0.24 mgm. of K_2O in set 3 increased the ammonia formation 19.6 mgm. over that in set 4. In comparing set 4 with the check, a slight increase in ammonia formation is evident when magnesium sulfate, calcium phosphate and no potassium sulfate were added. This amount, however, is practically

within experimental error. As is seen from an analysis of the peptone, 0.1 mgm. of potash was supplied with the dialyzed peptone, but this amount was sufficient to cause a production of only 8.7 mgm. of NH_3 expressed as nitrogen even after the other two salts were added. However, when a very small amount of this salt is added, a very great increase in activity is realized. Such minute amounts suffice probably because these elements are utilized again and again during the various successive cycles of the organisms.

TABLE 2

Potassium requirements for microorganisms as determined by the amounts of ammonia formed from dialyzed peptone by Bacillus subtilis

SET NO.	SALT PROPORTION	ACTUAL AMOUNTS OF K_2O ADDED	TOTAL CONCENTRATION OF SALT SOLUTION	AMMONIA FORMED	AMMONIA	
					Increase due to salts added	Increase due to K_2O
		mgm.	*atm.*	*mgm. N*	*mgm. N*	*mgm. N*
1	$1MgSO_4$ $1K_2SO_4$ $8Ca(H_2PO_4)_2.2H_2O$	1.25	0.100	29.9	23.6	21.2
2	$1MgSO_4$ $0.5K_2SO_4$ $8Ca(H_2PO_4)_2.2H_2O$	0.62	0.095	30.3	24.0	21.6
3	$1MgSO_4$ $0.2K_2SO_4$ $8Ca(H_2PO_4)_2.2H_2O$	0.24	0.092	28.3	22.0	19.6
4	$1MgSO_4$ $0K_2SO_4$ $8Ca(H_2PO_4)_2.2H_2O$	0.00	0.090	8.7	2.4	
Check (peptone alone)				6.3		

SUMMARY

With the physical conditions controlled, the results of the experiments here recorded indicate that:

1. By keeping the salts in the same proportion, the total concentration of a 3-salt solution can be reduced from 0.10 to 0.09 atmosphere without influencing the extent of ammonia formation from dialyzed peptone by *Bacillus subtilis*.

2. Potassium is essential to the development and activities of *Bacillus subtilis*.

3. If sufficient potassium for microorganisms is not present, magnesium sulfate and calcium phosphate will not influence the activities of (cannot be utilized by) *Bacillus subtilis*.

4. Supplying 0.24 mgm. of K_2O in addition to 0.1 mgm. already supplied in the dialyzed peptone in 100 cc. of solution, furnished sufficient potassium for the maximum development and activity of *Bacillus subtilis*. Applying two and one-half or five times this amount of potash did not increase the activity of the bacteria over that produced with 0.24 mgm.

REFERENCES

(1) Gerlach, M., and Vogel, I. 1903 Weitere Versuche mit Stickstoff-bindenden Bakterien. *In* Centbl. Bakt. (etc.), Abt. 2, Bd. 10, p. 636–643.

(2) Koch, George P. 1917 The effect of potassium salts upon biological activities. A thesis submitted to the faculty of Rutgers College in partial fulfillment of the requirements for the degree of Doctor of Philosophy, June, 1917.

(3) Koch, George P. 1917 Physiological balance in nutrient media for bacteria. Author's manuscript: not yet in print.

(4) Koch, George P. 1917 Effect of various salts upon the formation of ammonia by soil organisms. Paper presented at the meeting of the Biological-Chemical Section of the American Chemical Society at Kansas City, Mo., April 10, 1917.

(5) Krzemiemewska, Helen 1910 Influence of the mineral constituents of nutritive solutions on the development of *Azotobacter*. *In* Bul. Internat. Acad. Sci. Cracovie, Cl. Sci. Math. et Nat., Ser. B., p. 376–413.

(6) Liebig, Justus 1840 Chemistry in its Application to Agriculture and Physiology. Editions 1 and 2 (1840–1841).

(7) Nobbe, F., Schröder, J. and Erdmann, R. 1870 Über die organische Leistung des Kalium in der Pflanze. *In* Landw. Vers. Stat., Bd. 13, pp. 321–323.

(8) Shive, John W. 1915 A study of physiological balance in nutrient media. *In* Physiol. Res., v. 1, no. 7, p. 327–397.

(9) Tottingham, W. E. 1914 Quantitative chemical and physiological study of nutrient solutions for plant cultures. *In* Physiol. Res., v. 1, p. 133–245.

(10) Vogel, I. 1912 Untersuchungen über das Kalibedürfries von Azotobacter. *In* Centbl. Bakt. (etc.), Bd. 32, p. 411–420.

STUDIES ON THE CORRELATION BETWEEN THE PRODUCTION OF CARBON DIOXIDE AND THE ACCUMULATION OF AMMONIA BY SOIL ORGANISMS

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INTRODUCTION

The decomposition of organic matter in the soil being generally a biological process, various indexes of biological activity have been used to measure it, the more common of which have been plate counting, ammonification and nitrification. Wollny (14), Lemmerman and his associates (4), Stoklasa and Earnest (10), van Suchtelen (12), and others have shown that the evolution of carbon dioxide is an excellent index of the decomposition of soil organic matter. Later work has been done by Fred and Hart (2) and by Potter and Snyder (9) concerning the influnce of sulfates, phosphates and lime on carbon-dioxide production. Ammonification studies have been numerous but a survey of the literature yields little on the relationship between ammonia accumulation and carbon-dioxide production.

Discrepancies have been observed between an apparent increased biological activity in soils and an increased ammonia accumulation or vice-versa. Lipman and his associates (6) found that the addition of soluble carbohydrates decreased the accumulation of ammonia, and, in later work (7), of nitrates also, the production of carbon dioxide, however, being increased. It is evident that ammonia accumulation in itself can seldom be taken as a criterion of the intensity of action taking place. While ammonia is readily assimilated by most microorganisms and may therefore disappear as soon as liberated, the carbon dioxide produced is not used again by the same or by other species. Since carbon dioxide is not assimilated it becomes very nearly an absolute index of biological activity, with due regard to possible chemical formation under certain conditions.

In view of the fact that ammonia determinations are much more easily made and because much valuable ammonification data have been obtained, an attempt is being made to learn more of the relationship between the assimilation of ammonia and the actual activity as shown by the production of carbon dioxide. There must be an optimum ratio between these two factors necessary for the most favorable soil conditions.

Considerable attention is given here to the activity of pure cultures of some of the more common species of soil organism, both bacteria and fungi. Work with a fresh soil or soil infusions involves so many factors and species that it is difficult to interpret the results obtained, or at least to know the real effect or effects resulting from changing some of the conditions of the experiment. Results obtained with pure cultures and with mixtures of the same in the natural soil medium, except as it is changed by sterilization, ought to be of value in interpreting the action of the natural flora and to aid, ultimately, in the management and improvement of soils so that they will yield the greatest possible returns without deterioration.

Without at this time giving a more complete historical review there is here given a description of methods and of the apparatus, together with some of the data so far obtained.

THE APPARATUS AND ITS MANIPULATION

Some of the chief difficulties in obtaining a satisfactory apparatus for the determination of carbon dioxide evolved biologically, have been the lack of a source of slow, though continuous aspiration, too high a vacuum within certain of the parts, incomplete removal of the carbon dioxide from the air entering, incomplete absorption of the same gas produced, leakage, clogging of tubes, back suction and inconvenience in manipulation. It is believed that the apparatus used in this work does away with many of these defects although it may yet be much improved. Figure 1 gives a diagram of the different parts, and a photograph of a few of the units is given in plate 1. Air enters the bottle *A* containing strong sodium or potassium hydroxide and passes through the tower of glass beads inserted into the bottle. Most of the carbon dioxide and much of the moisture is removed in this tower which lies at an angle which may be varied to suit conditions. The air then passes through the soda-lime tube *B* for the more complete removal of carbon dioxide. From thence it enters the distributing bottle *C*, bubbling through a dilute acid, in this case 10 per cent sulfuric acid, before entering the bell-jar *D*. The bell-jar stands on a pine or cypress board which is prepared by painting on both sides with hot paraffin wax. A hole large enough for the insertion of a no. 5 2-holed rubber stopper is bored beneath each bell-jar which is made air-tight against the paraffined board by means of a brush and molten paraffin. The paraffin thus used was softened somewhat by melting it up with one-third its weight of paraffin oil. The air, drawn from the bottom of the bell-jar, passes in through the side neck of the suction flask and up through the barium hydroxide bead tower *E*, which removes the carbon dioxide produced by the organisms in flask *F*. From thence the line leads to the suction pump *G* which is fed by a siphon from the constant-level water tank *H*.

In the apparatus as used, a bell-jar, tower and distributing bottle are connected up in sets of six, each leading back to one soda-lime tube and ahead

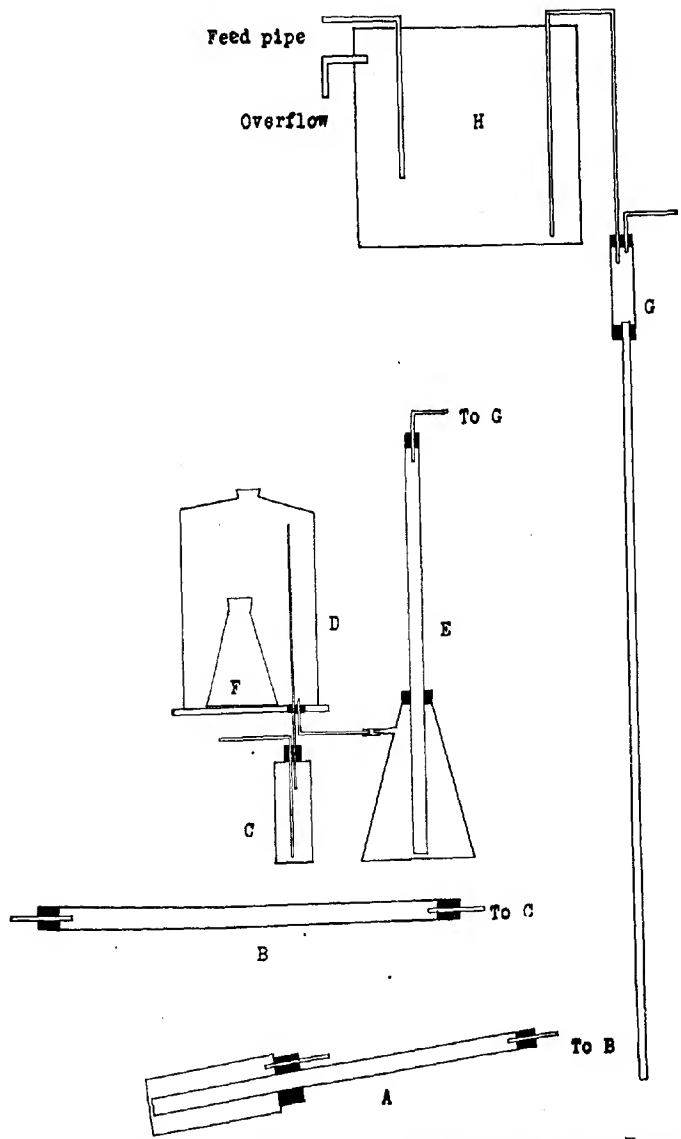

FIG. 1. A DIAGRAM OF THE APPARATUS USED TO DETERMINE CARBON DIOXIDE EVOLVED FROM SOIL[1]

[1] The principle involved in the pump was obtained from apparatus used by Dr. T. J. Headlee, entomologist of the New Jersey Agricultural Experiment Station.

to one suction pump. Each bell-jar and tower is in a separate line, however, diffusion of carbon dioxide being prevented by the acid solutions of the distributing bottles *C*. The suction pump *G* as finally constructed produced a very constant, continuous suction and required but little water for its operation. The tube leading from it being larger than the intake tube, bubbles of air are drawn down by the water in its descent. The pump itself consists of an ordinary thick-walled test-tube, fitted with stoppers at top and bottom, the closed end of the tube being broken off. The length of the drain tube largely determines the suction power and it was found that a tube 4 to 5 feet long was sufficient.

Clogging of the air inlet by the formation of crystals of sodium carbonate in the strong sodium hydroxide solution was prevented by using a bead tower *A* as shown, the opening at the bottom of the tower being 24 mm. in diameter. This tower was put in a slanting position to permit a slow, constant intake of air and to prevent as much of a vacuum as possible within the bell-jars.

The barium hydroxide tower which is 24 inches high is a modification of one described by Truog (11). The side-necked suction flask is of the type ordinarily used for Gooch crucible work. The barium hydroxide solution, used to absorb the carbon dioxide liberated, was made by adding 40 gm. of $Ba(OH)_2 8H_2O$ per liter of distilled water. The solution was siphoned off after standing over night and kept in a bottle protected from the carbon dioxide of the air, delivery being made through an automatic burette. The oxalic acid solution, used to titrate the excess of barium hydroxide, contained 8.6 gm. of the acid per liter. Both solutions were standardized in terms of milligrams of carbon dioxide per cubic centimeter. The procedure consisted of first adding the desired amount of barium hydroxide solution to the side-necked flask together with a few drops of phenolphthalein as an indicator and enough carbon-dioxide-free water to cause the liquid to rise about half way up in the bead tower when aspirating through the apparatus. When ready to make a determination the tower was partly withdrawn and washed free from barium hydroxide with carbon-dioxide-free water, the glass beads remaining in place because of the cloth gauze stretched over the bottom of the tube. The flasks were kept stoppered until titrated with oxalic acid, the residual barium hydroxide being thus determined. This method, although expeditious, results in some carbon-dioxide absorption from the air. Tests were conducted by aspirating for the customary periods with sterile flasks in the bell-jars, and the average blank thus obtained was subtracted from each determination.

The construction of the apparatus is such as absolutely to prevent any back suction of solution through the various units leading to the bell-jars. The amount of air aspirated was controlled with screw pinch cocks and was slow but continuous. Previous to a determination the air was allowed to circulate more rapidly for an hour. In view of the unquestioned value of carbon-dioxide determinations, it is hoped that a set of apparatus may finally

be perfected that will become as permanent a piece of laboratory equipment as an ammonia or a nitrogen still. Instead of bell-jars, for instance, metal jars, closed at the bottom and fitted with air-tight, removable covers, might be used.

METHODS

The bacteria used were grown on bouillon agar slants made in square bottles about 4 cm. wide and 10 cm. high. These were incubated at 28°C. for from 4 to 6 days. When ready to use, 100 cc. of a sterile 0.6 per cent salt solution was added to each bottle. A sterile rubber stopper was then inserted, and a suspension obtained by shaking gently. Erlenmeyer flasks of 250 cc. capacity, containing 70 gm. of soil were sterilized at 15 pounds steam pressure for 20 minutes and inoculated with 3 cc. of the suspensions. When mixtures of pure cultures were used for inoculating, these were obtained by pipetting equal amounts of the desired suspensions into a sterile flask from which 3-cc portions were drawn after gently shaking the mixture. This method of mixing reduces the total number of a given species but the inoculation was so heavy, at any rate, that this decrease was considered negligible.

When fungus cultures were employed the procedure was similar, except that a longer incubation of the inoculating material was necessary to secure abundant sporulation. The medium used consisted of 0.25 gm. of magnesium sulfate, 0.25 gm. of di-potassium phosphate, 10 gm. of peptone, 20 gm. of glucose, 15 gm. of agar and 1000 cc. of water. The soil infusions used were obtained from a neutral loam well supplied with organic matter.

The pure cultures employed were carefully tested as to purity and characteristics and were obtained from the stock cultures of the soil department of the New Jersey Agricultural Experiment Station. Several tests made upon flasks of soil which had been inoculated in the above manner showed no contamination with foreign organisms.

The soil was mixed with the organic material and other ingredients added by shaking for 5 minutes in a large bottle. The 70-gm. portions were then weighed out with a balance sensitive to 0.1 gm. The water content was 50 per cent of the maximum as determined by the Hilgard method. Seventy-gram instead of 100-gm. portions were used because a thinner layer with the same surface area was obtained, thus permitting better aeration. In the work done so far a soil classified as Norfolk sandy loam has been used. This contains sand of fine texture and has a maximum water-holding capacity of 32 per cent. It is low in organic matter, containing only 0.02 per cent of nitrogen, and has a lime requirement of 550 pounds of calcium oxide per acre as determined by the Veitch method.

Preliminary tests showed that the ammonia accumulation was about the same whether flasks were incubated without bell-jars or within, through which air was slowly but continuously circulated. The loss of moisture for a 12-day period was 1.2 gm. per flask within the bell-jars and 2 gm. without. Ac-

cordingly, some of the flasks for ammonia determinations were incubated apart from the apparatus during the same period that carbon-dioxide determinations were being made. The ammonia was determined by adding an excess of magnesium oxide and distilling in the usual way.

In all cases checks have been subtracted from the reported amounts of carbon dioxide and of ammonia, which are the averages of duplicate determinations. The inoculated flasks were incubated in a room kept at a temperature of from 21° to 23°C. for a period of 12 days.

TABLE 1

A comparison of duplicate determinations of carbon dioxide production and of ammonia accumulation

ORGANISMS	CO_2 PRODUCTION				NH_3 ACCUMULATION			
	1	2	Average	Per cent of error from mean	1	2	Average	Per cent of error from mean
Norfolk sandy loam + 1 per cent of cottonseed meal								
	mgm.	*mgm.*	*mgm.*		*mgm.*	*mgm.*	*mgm.*	
B. subtilis	134.1	138.8	136.5	3.4	10.38	11.40	10.59	9.5
B. vulgatus	169.1	167.0	168.1	1.3	7.85	8.61	8.23	9.2
B. subtilis + B. vulgatus	215.3	201.2	208.3	6.7	10.38	11.00	10.69	3.0
B. mycoides	60.4	Lost			2.00	2.27	2.19	12.3
B. megatherium	45.3	37.7	41.5	18.3	1.17	1.67	1.42	35.0
B. mycoides	40.7	43.5	41.6	4.1	1.42	1.42	1.42	00.0
B. mycoides + B. megatherium	38.4	39.6	39.0	3.0	2.82	2.32	2.57	2.0
B. mycoides + B. vulgatus	194.2	202.3	198.3	4.0	8.00	7.40	7.70	7.7
Norfolk sandy loam + 1 per cent of alfalfa meal								
B. subtilis	89.5	88.4	89.0	1.2	2.38	2.26	2.32	5.2
B, subtilis (no. P_2O_5 added)	81.7	79.9	80.8	2.2	2.51	2.51	2.51	0.0
B. megatherium	73.9	71.9	72.9	2.7	1.12	1.37	1.25	20.0
B. megatherium (no. P_2O_5 added)	63.4	60.1	61.8	5.3	1.37	1.24	1.31	10.0
Trichoderma sp.	160.1	146.1	153.1	9.1	0.69	0.57	0.63	*
Trichoderma sp. + Aspergillus niger	167.1	170.2	168.7	1.8	−0.32	−0.07	−0.20	
Aspergillus niger	145.2	138.9	142.1	4.4	−0.32	−0.32	−0.32	
Zygorhynchus Vuil + Trichoderma	133.2	133.1	133.2	0.0	0.44	0.57	0.52	
Zygorhynchus Vuil	104.6	90.5	97.6	14.4	0.95	0.95	0.95	
Soil infusion	253.8	272.4	263.1	7.1	−0.45	−0.45	−0.45	
Average				5.2				9.5

*These ammonia determinations from the fungus cultures were not included because of the small amounts obtained.

A COMPARISON OF THE DUPLICATE DETERMINATIONS OF CARBON-DIOXIDE PRODUCTION AND OF AMMONIA ACCUMULATION

It was observed that the variations between duplicate determinations of carbon dioxide were greater during the first few days. But at the end of 12 days the differences became less. Table 1 gives these duplicate determinations for the organisms discussed in the following pages. Averaging the percentages of error for each set of duplicates it was found that the average error for the carbon dioxide determinations was 5.2 per cent, while that for the ammonia determinations was 9.5 per cent. This indicates that carbon-dioxide production may be fairly accurately measured and that it was more uniform than the ammonia accumulation.

TABLE 2

Carbon dioxide and ammonia produced from 1 per cent of cottonseed meal in Norfolk sandy loam

TIME	B. SUBTILIS		B. VULGATUS		B. MYCOIDES		B. SUBTILIS + B. VULGATUS	
	CO_2	NH_3	CO_2	NH_3	CO_2	NH_3	CO_2	NH_3
days	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*
1 and 2	17.7		27.3		13.4		26.9	
3 and 4	37.1	5.30	49.1	none	9.3	0.73	64.8	3.97
5	13.2		25.7		6.2		17.6	
6	13.4	7.53	18.0	2.69	5.6	1.18	32.4	6.88
7 and 8	18.1	8.73	15.9	3.49	5.8	1.88	27.0	9.37
9 and 10	22.8	10.58	23.0	4.86	8.1	1.56	24.6	10.00
11 and 12	14.2	10.89	9.1	8.23	12.0	2.19	15.1	10.69
Totals	136.5		168.1		60.4		208.4	
Per cent of total nitrogen and of total carbon as found	17.8	26.6	21.8	20.1	7.9	5.4	27.1	26.1

THE RELATIVE OXIDIZING AND AMMONIFYING POWER OF SEVERAL PURE CULTURES AND OF MIXTURES OF THE SAME

One per cent of cottonseed meal was the source of organic matter for the series given in table 2. It contained 5.84 per cent of nitrogen and 40.3 per cent of carbon. This was mixed with Norfolk sandy loam to which was added 0.066 per cent of acid phosphate, containing about 16 per cent of water-soluble phosphorus calculated as phosphoric pentoxide. Three-tenths of 1 per cent of precipitated calcium carbonate also was added. These percentages are equivalent to about 2000 pounds and 9000 pounds per acre 9 inches. Table 3 gives data obtained under similar conditions for another series.

Considering first the relative activities of these common soil types, it may be seen that *Bacillus vulgatus* leads in carbon-dioxide production (table 2), oxidizing, in a 12-day period, 21.8 per cent of the carbon added in the cotton-

seed meal. *Bacillus subtilis*, giving considerably less carbon dioxide, caused the accumulation of more ammonia, amounting to 26.6 per cent of the total nitrogen added. A mixture of these two organisms gave a higher carbon-dioxide production than either alone, and an ammonia accumulation approximating that of *B. subtilis*. *Bacillus mycoides*, tested twice (tables 2 and 3), gave very much lower amounts both of carbon dioxide and of ammonia. *Bacillus megatherium* (table 3) was about as active as *B. mycoides* and a mixture of the two gave no increase in carbon dioxide. But mixing *B. vulgatus* with *B. mycoides* caused a very marked increase both in carbon-dioxide production and in ammonia accumulation. This increase in carbon dioxide is easily observed in figure 3. Both figure 2 and figure 3 show that the maximum rate of carbon-dioxide production always occurred before the fifth day of incubation.

TABLE 3

Carbon dioxide and ammonia produced from 1 per cent of cottonseed meal in Norfolk sandy loam

TIME	B. MYCOIDES		B. MEGATHERIUM		B. MEGATHERIUM + B. MYCOIDES		B. VULGATUS + B. MYCOIDES	
	CO_2	NH_3	CO_2	NH_3	CO_2	NH_3	CO_2	NH_3
days	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*
1 and 2	12.9		13.8		11.6		36.1	
3	3.0	0.50	3.9	1.19	2.0	0.75	20.6	0.63
4 and 5	5.6	1.45	8.6	1.32	4.8	1.51	43.1	4.81
6, 7, 8 and 9	11.5		9.8		14.8		75.1	
10	2.6		1.7		1.8		8.8	
11 and 12	5.8	1.42	3.7	1.42	4.1	2.57	14.7	7.70
Totals	41.6		41.5		39.1		198.4	
Per cent of carbon and of nitrogen as found	5.4	3.5	5.2	3.5	5.0	6.3	25.8	18.8

A series in which ground alfalfa hay was used instead of cottonseed meal was inoculated with cultures of *B. subtilis* and *B. megatherium* (table 4). As shown in the table, the addition of acid phosphate caused an increase in carbon-dioxide but none in ammonia accumulation. Until more data are secured no conclusions can be drawn in the comparison of carbon-dioxide production in its relation to ammonia accumulation as influenced by phosphates and by other salts.

In a second series, with alfalfa as the source of organic matter, the activities of some fungi were measured and compared with those of a soil infusion (table 5). The average oxidizing power for the bacteria (table 4) was 8.0 per cent, while that for the individual species of fungi was 16.5 per cent of the carbon added. The soil infusions greatly exceeded both, being 33.0 per cent. The bacteria are not able to utilize the carbon of cured alfalfa as readily as fungi,

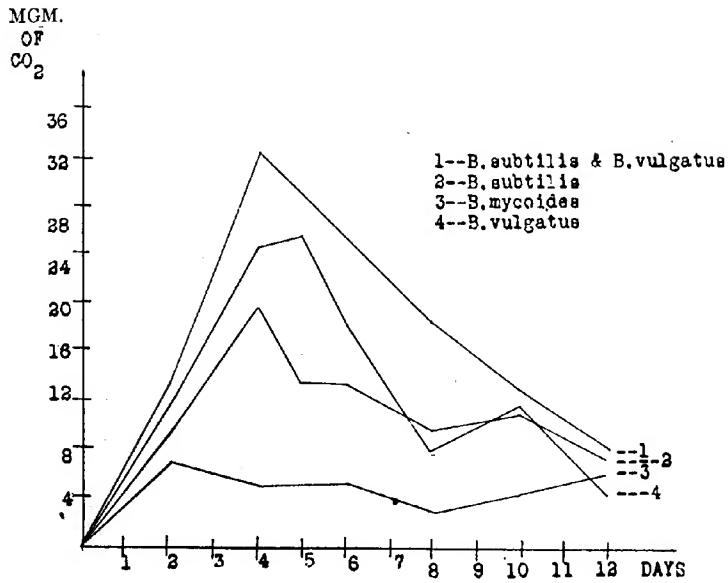

FIG. 2. THE DAILY PRODUCTION OF CARBON DIOXIDE FROM NORFOLK SANDY LOAM, PLUS 1 PER CENT OF COTTONSEED MEAL, BY PURE CULTURES OF BACTERIA (TABLE 2)

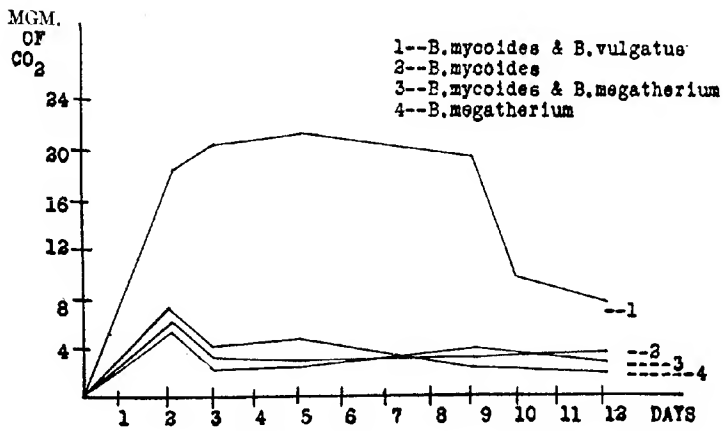

FIG. 3. THE DAILY PRODUCTION OF CARBON DIOXIDE FROM NORFOLK SANDY LOAM, PLUS 1 PER CENT OF COTTONSEED MEAL, BY PURE CULTURES OF BACTERIA (TABLE 3)

and this would be even more evident for a longer period, since the fungus cultures start more slowly but are more active at the end of 12 days.

Glancing again at table 5 it may be seen that when a mixed culture of *Trichoderma* sp. and *Aspergillus niger* is used an activity greater than that of either alone is obtained. *Trichoderma* sp. and *Zygorhychus Vuilleminii* acting together showed an activity less than the one and greater than the other. It would be interesting to know more of the associative action of fungi and bacteria in pure cultures. The combinations of bacterial cultures so far studied, appear to be symbiotic in their behavior. Using ammonia accumulation as an index, Coleman (1) found, for low moisture contents, an antagonistic action between *B. subtilis* and *Zygorhynchus Vuilleminii*. Soil

TABLE 4

Carbon dioxide and ammonia produced from 1 per cent of alfalfa in Norfolk sandy loam with and without the addition of acid phosphate

TIME	B. SUBTILIS				B. MEGATHERIUM			
	Acid phosphate added		No acid phosphate added		Acid phosphate added		No acid phosphate added	
	CO_2	NH_3	CO_2	NH_3	CO_2	NH_3	CO_2	NH_3
days	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*
1	7.1		5.7		7.8		7.4	
2 and 3	41.6		38.6		30.0		26.6	
4	13.0		8.3		7.4		5.0	
5 and 6	12.6	1.80	11.2	1.63	10.2	1.11	9.8	0.73
7 and 8	6.1		6.6		6.8		4.7	
9, 10, 11 and 12	8.7	2.32	10.1	2.51	10.9	1.25	8.3	1.31
Totals	89.0		80.8		72.9		61.8	
Per cent of carbon and of nitrogen as found	8.4	14.9	6.8	16.1	7.6	8.0	5.8	8.4
Per cent increase due to acid phosphate	10.1				17.9			

infusions are known to be more active than individual organisms. Hence it may be inferred either that the associative action of the many species in an infusion is beneficial or that the most active species have not been isolated, and if they have, that they have been attenuated by artificial culture.

Finally, it may be observed (table 5) that those fungi which oxidized the most gave the lowest accumulations of ammonia. In both cases where *Aspergillus niger* was added, as well as where a soil infusion was employed, the organisms even used up some of the ammonia shown to be present in the sterilized checks (fig. 5).

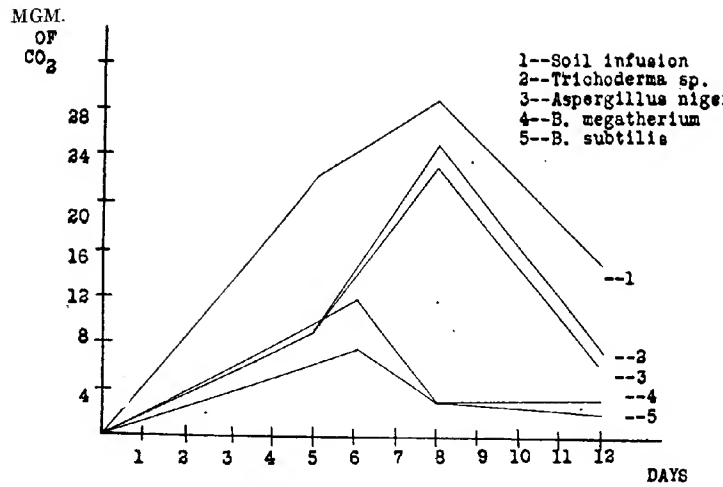

FIG. 4. THE PRODUCTION OF CARBON DIOXIDE BY CULTURES OF BACTERIA AND OF FUNGI AND BY A SOIL INFUSION, WITH 1 PER CENT OF CURED ALFALFA IN NORFOLK SANDY LOAM (TABLES 4 AND 5)

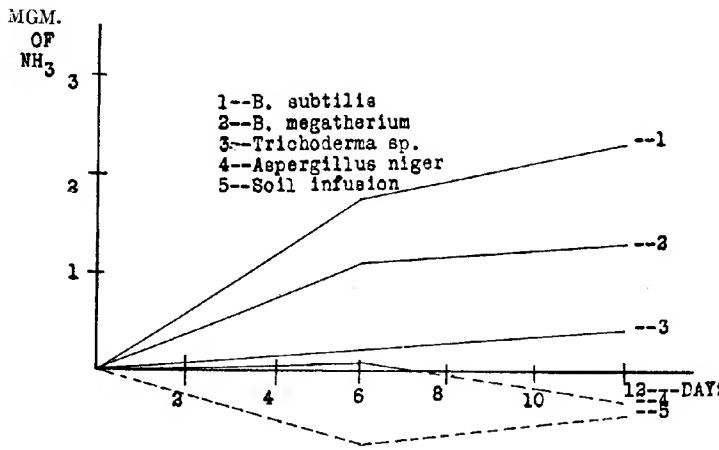

FIG. 5. THE PARALLEL ACCUMULATION OF AMMONIA BY THE ORGANISMS FOR WHICH THE CARBON-DIOXIDE PRODUCTION IS GIVEN IN FIGURE 4

THE CORRELATION BETWEEN CARBON-DIOXIDE PRODUCTION AND AMMONIA ACCUMULATION BY PURE CULTURES OF BACTERIA AND FUNGI AND BY SOIL INFUSIONS

In considering the relation of ammonia accumulation to the rate of organic decay it is of interest to inspect the data so far obtained from parallel determinations of ammonia and carbon-dioxide. Table 6 shows that a soil infusion decomposed nearly three times as much of the alfalfa as *B. subtilis* for a period of 8 days, the percentages of total carbon oxidized being 22.0 and 8.9, respectively. During this period *B. subtilis* caused the accumulation of 10.4 per

TABLE 5

Carbon dioxide and ammonia produced from 1 per cent of alfalfa in Norfolk sandy loam by fungi and by a soil infusion

TIME	TRICHODERMA SP.		ASPERGILLUS NIGER		ZYGORHYNCHUS VUILLEMINII		SOIL INFUSION		TRICHODERMA SP. + ASPERGILLUS NIGER		TRICHODERMA SP. + ZYGORHYNCHUS VUILLEMINII	
	CO_2	NH_3	CO_2	NH_3	CO_2	NH_3	CO_2	NH_3	CO_2	NH_3	CO_2	NH_3
days	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*
1, 2, 3, 4 and 5	46.0	0.04	47.5	0.13	38.0	0.63	112.8	−0.72	45.4	0.45	38.6	0.31
6, 7 and 8	77.6	0.25	70.1	0.00	44.6	0.44	88.3	−0.22	91.0	−0.14	59.8	0.70
9, 10, 11 and 12	29.6	0.63	24.5	−0.32	15.1	0.95	62.1	−0.45	32.3	−0.20	34.9	0.52
Totals.....	153.2		142.1		97.7		263.1		168.7		133.3	
Per cent of carbon and of nitrogen as found.	19.4	4.4	17.7		12.3	6.1	33.0		21.1		16.7	3.4

TABLE 6

The production of carbon dioxide and the accumulation of ammonia from 2 per cent of alfalfa in Norfolk sandy loam by B. subtilis and a soil infusion

ORGANISMS	1ST DAY	2ND DAY	3RD DAY	4TH DAY	5TH AND 6TH DAYS	7TH AND 8TH DAYS	TOTALS	PER CENT OF CARBON OXIDIZED	PER CENT OF N AMMONIFIED
	mgm.	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*		
B. subtilis									
CO_2.....	47.3	39.6	16.7	9.8	15.6	11.2	140.2	8.9	
NH_3.....		1.70		2.14	3.13	3.49			10.4
Soil infusion									
CO_2.....	47.0	42.0	68.2	54.6	91.8	41.8	345.4	22.0	
NH_3.....		−0.60		−0.51	−1.20	−1.16			none

cent of the total nitrogen as ammonia, whereas the soil infusion not only caused no accumulation but even used up most of the small amount appearing in the checks after sterilizing.

With alfalfa as the source of energy pure cultures of fungi give results similar to those obtained with a soil infusion (table 5). The less active of the fungi tested gave a small accumulation of ammonia while the more active species, like the soil infusions, used up some of the ammonia originally present in the checks as well as all that may have been liberated.

Looking now at the action of bacteria upon alfalfa it may be noted that they (table 4) cannot utilize it as readily as fungi, the average amount oxidized by the two groups being 8.0 and 17.5 per cent, respectively. But the bacteria, although they oxidized less than half as much carbon, caused the accumulation of 14.2 per cent of the total nitrogen as ammonia, or over three times as much as was accumulated by the fungi, for which the percentage was 4.6, not including the more active fungus cultures which produced no ammonia at all (fig. 4 and 5).

Under the conditions of the experiment, an average of 12.2 per cent of the total carbon of cottonseed meal was oxidized by the bacterial cultures (tables 2 and 3), while the average amount of nitrogen found as ammonia was 13.8 per cent. The action of fungi upon cottonseed meal has not been studied with reference to the simultaneous production of carbon dioxide and accumulation of ammonia. Working with pure cultures, McLean and Wilson (8), and later Coleman (1), Kopeloff (3), and Waksman and Cook (13), have shown that fungi cause considerable accumulation of ammonia from cottonseed meal, the amounts depending upon various factors such as phosphates, length of incubation, moisture, temperature and type of soil. Lipman and Burgess (5), studying several bacteria in pure cultures, found with a given type of soil, that the species giving the highest ammonia accumulation with one kind of organic matter was not always the highest accumulator with another kind.

Referring again to the carbon-dioxide production and the ammonia accumulation from alfalfa, the data so far secured show that a low accumulation of ammonia is an indication of high rather than of low activity. With this type of organic matter the behavior of fungi was more like the action of soil infusions than the behavior of bacteria, indicating that the most active components of a soil flora, as obtained from an infusion, are fungi.

It seems evident that other kinds of organic matter and of soils, as well as the effect of various fertilizing elements and soil amendments, should be studied with reference to the optimum ratio between carbon-dioxide production and ammonia accumulation. The action of pure cultures of bacteria, actinomyces and fungi, when mixed together in different combinations, deserves further consideration and investigation. If these three groups make up the natural soil flora, studies in such combinations or floras made up synthetically might throw more light on the interactions and needs of soil organ-

isms so that they could be better controlled and aided in their function of preparing food for plants.

SUMMARY

The results obtained so far with the use of carbon dioxide as an index of biological activity and the correlation of this activity with ammonia accumulation may be summarized as follows.

1. An apparatus is described for the determination of carbon dioxide evolved biologically from soil.

2. Methods are given which were used for the study of organic decomposition by pure cultures and by mixtures of pure cultures of microorganisms.

3. Duplicate determinations indicate that the production of carbon dioxide is more uniform than the accumulation of ammonia for a 12-day period.

4. In general a high carbon-dioxide production by pure cultures of bacteria was accompanied by a high ammonia accumulation, with 1 per cent of cottonseed meal or of alfalfa in Norfolk sandy loam.

5. Pure cultures of the fungi tested, oxidized more of the carbon of alfalfa than pure cultures of bacteria, but the bacteria caused the accumulation of much more ammonia. The more active species of fungi not only caused no accumulation of ammonia but even used up some of the small amounts appearing in the checks. Soil infusions resembled the fungus cultures with respect to ammonia accumulation but were more active in the production of carbon dioxide.

6. With alfalfa as the source of organic matter a low accumulation of ammonia is an indication of a high rather than of a low activity. Furthermore, since the behavior of the soil infusions was more like that of fungi than of bacteria, it would seem that fungi were the more active components of the natural soil flora.

7. The mixtures of pure cultures of bacteria tested showed no antagonism and in some cases a symbiotic relation seemed to exist. Mixtures of pure cultures of fungi or of fungi and bacteria have not been studied sufficiently to permit of any conclusion therefrom.

In conclusion it is a pleasure to thank Dr. J. G. Lipman for his many helpful suggestions given during the course of this work.

REFERENCES

(1) Coleman, D. A. 1916 Environmental factors influencing the activity of soil fungi. *In* Soil Sci., v. 2, no. 1, p. 1–65.

(2) Fred, E. B., and Hart, C. B. 1915 The comparative effect of phosphates and sulfates on soil bacteria. Wis. Agr. Exp. Sta. Research Bul. 35, 32 p.

(3) Kopeloff, N. 1916 The inoculation and incubation of soil fungi. *In* Soil Sci., v. 1, no. 4, p. 381–403.

(4) Lemmermann, O., Aso, K., Fischer, H., and Fresenius, L. 1911 Untersuchungen über der Zersetzung der Kohlenstofferbindungen verschiedener organischer Substanzen im Boden, speziell unter dem Einfluss von Kalk. *In* Landw. Jahrb., Bd. 41, p. 217–256.

(5) Lipman, C. B., and Burgess, P. S. 1914 Ammonification in soils by pure cultures. *In* Univ. Cal. Pub. Agr. Sci., v. 1, no. 7, p. 141–172.

(6) Lipman, J. G., Blair, A. W., Owen, I. L., and McLean, H. C. 1912 Experiments on ammonia formation in the presence of carbohydrates and other non-nitrogenous organic matter. N. J. Agr. Exp. Sta. Bul. 247, 22 p.

(7) Lipman J. G., and Blair, A. W. 1914 The formation of carbon dioxide and nitrates in the presence of large amounts of carbohydrates. *In* N. J. Agr. Exp. Sta. 35th Ann. Rpt., 1914, p. 220–221.

(8) McLean, H. C., and Wilson, G. W. 1914 Ammonification studies with soil fungi. N. J. Agr. Exp. Sta. Bul. 270, 39 p.

(9) Potter R. S., and Snyder, R. S. 1916 Carbon dioxide production in soils and nitrogen changes in soils variously treated. Iowa Agr. Exp. Sta. Research Bul. 39, p. 254–309.

(10) Stoklasa, J., and Earnest, A. 1905 Über den Ursprung, die Menge und die Bedeutung des Kohlen dioxyds im Boden. *In* Centbl. Bakt. [etc.], abt. 2, Bd. 14, p. 723–736.

(11) Truog, E. 1915 Methods for the determination of carbon dioxide and a new form of absorption tower adapted to the titrimetric method. *In* Jour. Indus. Engin. Chem., v. 7, no. 12, p. 1045.

(12) van Suchtelen, F. H. H. 1910 Über die Messung der Lebenstätigheit der äerobiotischen Bakterien im Boden durch die Kohlensäureproduktion. *In* Centbl. Bakt. [etc.], Abt. 2, Bd. 14, p. 45–89.

(13) Waksman, S. A., and Cook, R. C. 1916 Incubation studies with soil fungi. *In* Soil Sci., v. 1, no. 3, p. 275–284.

(14) Wollny, E. 1897. Die Zersetzung der Organischen Stoffe.

PLATE 1

A part of the carbon-dioxide apparatus set up in the constant temperature room. (Photo by R. F. Poole.)

EXPERIMENTS WITH SULFUR-PHOSPHATE COMPOSTS CONDUCTED UNDER FIELD CONDITIONS[1]

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SERIES I. COMPOSTS IN EARTHENWARE POTS

It is often advantageous in research work to imitate natural conditions without sacrificing laboratory precision and careful control. Such at least was the need felt in carrying out the sulfur-floats composting experiments. It was desired to have certain mixtures placed in sufficiently direct contact with the soil to create natural conditions of soil moisture and temperature and yet to retain the mixtures well defined, thus assuring representative sampling, and also artificial control of atmospheric moisture. This requirement was met by the use of porous earthenware pots—each provided with a hole in the bottom—placed in slight excavations, which were so adjusted that the tops of the pots were flush with the surface of the soil.

The experiment as already stated was to deal with a variety of conditions rather than with varying combinations of materials.

Four standard mixtures were used, viz.,

No. 1. Red silt loam soil (0.1190 per cent P_2O_5).
No. 2. 2 parts sulfur to 12 parts soil.
No. 3. 6 parts floats to 12 parts soil.
No. 4. 6 parts floats, 2 parts sulfur, and 12 parts soil.

Four sets of these mixtures were made up—the proportions having been so adjusted that there were 20 pounds in each pot. Each set was placed under a different condition. Two sets were covered with waterproof canvas placed on a V-shaped frame open at each end. The other two sets were entirely exposed to the weather. The arrangement of the pots and the treatment which each received were as follows:

[1] See Lipman, J. G., and McLean, H. C. The oxidation of sulfur in soil as a means of increasing the availability of mineral phosphates. *In* Soil Sci., v. 1, p. 533, (1916). Sulfur oxidation in soils and its effect on the availability of mineral phosphates. *In* Soil Sci., v. 2, p. 499 (1916). Vegetation experiments on the availability of treated phosphates. *In* Soil Sci., v. 4, p. 337 (1917).

1. Soil, compost covered, inoculated.
2. Soil, sulfur, covered, inoculated.
3. Soil, floats, covered, inoculated.
4. Soil, floats and sulfur, covered, inoculated.
5. Soil, uncovered, 0.02 per cent aluminum sulfate, ferrous sulfate, inoculated.
6. Soil, sulfur, uncovered, 0.02 per cent aluminum sulfate, 0.02 per cent ferrous sulfate, inoculated.
7. Soil, floats, uncovered, 0.02 per cent aluminum sulfate, 0.02 per cent ferrous sulfate, inoculated.
8. Soil, floats, sulfur, uncovered, 0.02 per cent aluminum sulfate, 0.02 per cent ferrous sulfate, inoculated.
9. Soil, covered, 0.02 per cent aluminum sulfate, 0.02 per cent ferrous sulfate, inoculated.
10. Soil, sulfur, covered, 0.02 per cent aluminum sulfate, 0.02 per cent ferrous sulfate, inoculated.
11. Soil, floats, covered, 0.02 per cent aluminum sulfate, 0.02 per cent ferrous sulfate, inoculated.
12. Soil, floats, sulfur, covered, 0.02 per cent aluminum sulfate, 0.02 per cent ferrous sulfate, inoculated.
13. Soil, uncovered, inoculated.
14. Soil, sulfur, uncovered, inoculated.
15. Soil, floats, uncovered, inoculated.
16. Soil, floats, sulfur, uncovered, inoculated.

The quantity of available phosphoric acid found in the composts at different intervals is recorded in table 1.

A discussion of the high production of available phosphoric acid where sulfur and floats were combined, as compared with the production in the other composts of each set, scarcely seems necessary. In view of the extensive data recorded elsewhere this was to be expected. Consideration will be given at this time only to the sulfur-floats combinations as affected by the environment to which they were exposed.

Passing over the steady increase of available phosphoric acid at the successive dates of sampling, and comparing the highest results obtained, which is at the end of 20 weeks, we note first of all that the cover protection exerted some influence. The pots under cover exhibit a higher available phosphoric acid yield than do the others. No. 4 shows an accumulation of 36.86 per cent of the total phosphoric acid in an available form, as against 29.68 per cent in No. 16. In the case of the composts which received the aluminum and iron salts we likewise observe a 32.55–32.96 per cent accumulation of available phosphoric acid. As the covered pots were kept under optimum moisture conditions by artificial means, while the uncovered pots were always exposed, it would be expected that a greater chance for leaching would develop there; especially so, since rainy weather prevailed during a considerable part of the first fifteen weeks of the experiment. It is to leaching, therefore, that we ascribe this difference.

The aluminum and iron salts did not, it seems, produce results like those obtained in laboratory experiments. In this instance the ferrous and aluminum sulfates brought about an increase only in the uncovered set, and

this increase is not very large. In compost 8, we find 62.40 pounds of available phosphoric acid, as against 56.20 pounds in compost 16, which received no addition of salts. In the covered set we find 69.80 pounds of available phosphoric acid in the compost receiving no salts as against 61.63 pounds in the treated ones. As the ferrous and aluminum sulfates were not added until 5 weeks after the experiment was started, it is probable that this circumstance might account for the discrepancies noted. In the laboratory experiments the salts were always added when the composts were being started and hence it is probable that a stimulation was exerted at the time when the sulfofiers were developing vigorously.

TABLE 1

The content of available P_2O_5 in the different composts calculated to pounds of P_2O_5 per ton

COMPOST NUMBER	AT BEGINNING	AETER 8 WEEKS	AFTER 11 WEEKS	AFTER 15 WEEKS	AFTER 20 WEEKS	TOTAL P_2O_5 IN 1 TON OF COMPOST	TOTAL P_2O_5 RENDERED AVAILABLE IN 20 WEEKS
	pounds	*pounds*	*pounds*	*pounds*	*pounds*	*pounds*	*per cent*
1	0.53	0.64	0.63	0.64	0.598	1.95	
2	0.53	0.63	0.64	0.61	0.60	1.67	
3	2.96	2.90	3.32	3.29	3.45	208.76	1.65
4	3.13	8.36	23.51	53.77	69.80	189.32	36.86
5	0.53	0.64	0.61	0.63	0.65		
6	0.53	0.63	0.64	0.58	0.60		
7	2.96	3.24	3.53	3.49	3.80	208.76	1.82
8	3.13	10.82	19.23	49.08	62.40	189.32	32.96
9	0.53	0.58	0.62	0.64	0.65		
10	0.53	0.63	0.64	0.61	0.71		
11	2.96	3.27	3.41	3.29	3.40	208.76	1.62
12	3.13	5.78	14.56	54.04	61.63	189.32	32.55
13	0.53	0.61	0.63	0.62	0.64		
14	0.53	0.63	0.62	0.64	0.78		
15	2.96	3.09	3.49	3.33	3.13	208.76	1.49
16	3.13	8.81	15.91	51.81	56.20	189.32	29.68

SERIES II. PRODUCTION OF AVAILABLE P_2O_5 IN 2-TON COMPOSTS

The theory of available phosphate production from insoluble phosphates, through the medium of sulfur, having been established by laboratory experimentation, the next step would seem to be its commercial utilization.

In the case at hand, the problem contains the practical application under farm conditions rather than from the factory standpoint. How can the individual farmer utilize the sulfur-floats method? This question can be best answered by actual field tests. Accordingly a number of composts of varying composition were made up in the latter part of October, 1916. The soil used was a red silt loam. It was sifted through a quarter-inch mesh sieve to facilitate handling. Tennessee brown rock phosphate containing 31.12

per cent of phosphoric acid, sulfur flour, and composted manure were the materials used in the composts. Samples of the manure and soil were secured and analyzed in order that the per cent of phosphoric acid rendered available in the compost could be calculated. The composts were made up so that at completion each weighed two tons. They were prepared according to the following plan:—

Compost No.	
1.	4,000 pounds soil (0.1190 per cent P_2O_5).
2.	3,800 pounds soil, 200 pounds sulfur.
3.	3,600 pounds soil, 400 pounds floats.
4.	3,400 pounds soil, 400 pounds floats, 200 pounds sulfur.
5.	3,600 pounds soil, 400 pounds sulfur,
6.	3,200 pounds soil, 800 pounds floats,
7.	2,800 pounds soil, 800 pounds floats, 400 pounds sulfur.
8.	3,800 pounds soil, 200 pounds manure.
9.	3,600 pounds soil, 200 pounds manure, 200 pounds sulfur.
10.	3,400 pounds soil, 200 pounds manure, 400 pounds floats.
11.	3,200 pounds soil, 200 pounds manure, 400 pounds floats, 200 pounds sulfur.
12.	3,400 pounds soil, 200 pounds manure, 400 pounds sulfur.
13.	3,000 pounds soil, 200 pounds manure, 800 pounds floats.
14.	2,600 pounds soil, 200 pounds manure, 800 pounds floats, 400 pounds sulfur.

In each compost a thorough mixing of the constituents was obtained by shovelling over several times. Moreover, to insure an even mixture the sulfur, where used, was sifted through a fine-mesh sieve before its addition. Fourteen composts were made and exposed to the weather. They were so spaced as to prevent contamination of one heap by another. They were turned over on May 1, 1917. At different intervals samples were secured and analyzed for citrate-soluble phosphoric acid. The dates of sampling, and the results obtained are given in table 2.

The maximum production of available phosphoric acid is shown at the end of 44 weeks. Since 400 pounds of rock phosphate contain 124.48 pounds of phosphoric acid, we at once note the high proportion made available in the composts containing both floats and sulfur. Likewise we note, in contrast, the small amounts of available phosphoric acid obtained where sulfur was not employed with the floats.

We note, further, that the check compost number 1 contained about the lowest amount of available phosphoric acid in its series, numbers 1 to 7; as also did the check compost number 8 for its series, numbers 8 to 14. Sulfur alone and with manure gave no appreciable increase over the check. In the case of the composts 3, 6, 10 and 13, which received floats, but no sulfur, it will be noted that the insoluble phosphates had undergone practically no changes in solubility throughout the entire 44-week period. It is only when the floats are composted with sulfur that a change in availability is effected. The remarkable increase in available phosphoric acid from May 12 to September 3, should be especially noted. It appears that the sulfofication proc-

ess did not gain headway until the advent of warm weather. Once started it apparently developed very rapidly, until, on September 3, we find 96.98 pounds, or 75.87 per cent of the total phosphoric acid, to be available in compost 4 where 400 pounds of floats, 200 pounds of sulfur, and 3400 pounds of soil were composted together. In compost 7 with the quantities of sulfur and floats doubled and the soil reduced to 2800 pounds, we note but a slight increase in available phosphoric acid over that obtained in compost 4. The amount of available phosphoric acid in this case was 99.79 pounds, or 39.65 per cent of the total. The addition of manure did not increase the accumulation of available phosphoric acid in compost 11, for only 96.43 pounds were obtained here. This amount is the same as that obtained in compost 4 which contained no manure. In compost 14, however, the addition of manure evidently favored sulfofication, because 174.38 pounds, or 69.16 per cent of

TABLE 2

The content of available phosphorus in the composts at various intervals calculated to pounds of P_2O_5

COMPOST NUMBER	AT BEGINNING OCTOBER 31, 1916	AFTER 10 WEEKS JANUARY 10, 1917	AFTER 21 WEEKS MARCH 30, 1917	AFTER 27 WEEKS MAY 12, 1917	AFTER 38 WEEKS JULY 23, 1917	AFTER 44 WEEKS SEPTEMBER 3, 1917	TOTAL P_2O_5 IN COMPOST	TOTAL P_2O_5 RENDERED AVAILABLE IN 44 WEEKS
	pounds	*pounds*	*pounds*	*pounds*	*pounds*	*pounds*	*pounds*	*per cent*
1	0.97	0.96	1.01	0.92	0.95	1.07		
2	0.93	0.95	1.05	1.03	1.02	1.11		
3	3.79	4.25	3.77	4.26	4.89	6.45	127.99	5.04
4	3.85	4.99	5.18	7.88	47.96	96.98	127.82	75.87
5	1.16	1.14	1.14	1.01	1.21	1.16		
6	6.09	4.85	4.85	4.48	6.95	7.51	252.08	2.98
7	5.65	6.50	6.89	8.99	49.99	99.79	251.69	39.65
8	1.27	1.57	1.34	1.36	1.16	2.22		
9	1.41	1.45	1.35	1.38	1.47	1.73		
10	3.36	2.35	3.68	3.33	4.86	5.23	127.80	4.09
11	3.93	4.38	4.83	9.61	56.01	96.43	127.60	75.85
12	1.78	1.50	1.63	1.69	1.61	2.89		
13	4.76	5.46	4.98	4.76	6.95	6.25	252.53	2.47
14	4.25	7.62	7.04	25.58	147.49	174.38	252.14	69.16

the total phosphoric acid, were found to be available in this case as against 99.79 pounds, or 39.65 per cent of the total phosphoric acid, in compost 7, which received the same quantities of sulfur and floats but no manure. On May 12, similar relative results were obtained. From October, 1916, to May 1917, sulfofication was practically suspended, although on May 1 it was observed that where sulfur was one of the constituents of the composts, their exterior, to a depth of one inch, showed an almost total absence of sulfur. This indicated that it had been transformed into some other form, that is, it had been oxidized. Hence it is likely that even during the cold months some action had taken place in the outer layer of the compost, which was influenced either by aeration or temperature.

In conclusion, it appeares that composts should be made up in warm weather, and so constructed as to permit free access of air, if maximum results are to be obtained.

SERIES III. SHALLOW COMPOSTS

Laboratory experiments showed aeration to be an important factor in composting. On May 1, when the composts in the experiment already reported were turned, it was observed that the outer layer to the depth of one inch was devoid of sulfur, indicating that the sulfur was transformed rapidly only in the well aerated outer layer. It was therefore concluded that large composts, especially when placed in large heaps, were not the most desirable. The need of thorough aeration having been indicated, it was decided to prepare composts with a depth of about 15 inches. This was on a small scale, the plots each being only 4 feet square. The soil, a Sassafras gravelly loam, was considered to weigh 80 pounds per cubic foot. With this soil contained

TABLE 3

Available phosphoric acid in shallow compost experiment calculated in pounds per ton of compost

PLOT NUMBER AND TREATMENT	AT BEGINNING	AFTER 6 WEEKS	AFTER 15 WEEKS	AFTER 20 WEEKS
	pounds	*pounds*	*pounds*	*pounds*
1. 320 pounds floats	3.49	3.38	3.89	3.26
2. 320 pounds floats and 106.66 pounds sulfur	3.98	4.96	27.39	35.41

within the 4 x 4 foot plot to a depth of 6 inches, the floats and sulfur were thoroughly mixed. Each compost was inoculated with soil known to possess a high sulfofying power. The composts were stirred every week, thus keeping them well aerated. The plan of the experiment and the results obtained are recorded in table 3.

Each ton of the composted material was calculated to contain 186.72 pounds of phosphoric acid in the floats alone, We therefore note with interest the contrast between the two composts, a low availability with the one; a high availability with the other. At the end of 6 weeks the sulfur-treated floats do not show such contrast, but after that the available phosphoric acid accumulated very rapidly until at the end of 20 weeks we find as much as 35.41 pounds of available phosphoric acid per ton of the air-dry compost as against 3.26 pounds in the compost containing only soil and floats. These results seem to indicate that the better aerated mixtures were more effective than the 2-ton composts, for even at the end of 6 months the latter did not show as great an accumulation of available phosphoric acid as was obtained in this experiment at the end of 15 weeks. One point should be remembered, the 2-ton composts were started in October at the beginning of cold weather, while this experiment was started at the beginning of warm weather (April 18). Nevertheless, this experiment disclosed many advantages existing in

the use of shallow composts, and further trials for this reason were made with composts described in the next series.

SERIES IV. MODIFIED SHALLOW COMPOSTS

This experiment was designed to ascertain the best depth through which sulfur and floats may be distributed in the soil, in order to obtain a maximum accumulation of available phosphoric acid. Five small plots, 2 feet square, were used. The soil within each area was removed to a different depth for each plot. The depths were; ½ inch, 1 inch, 2 inches, 3 inches, and 4 inches. The soils from the different plots were mixed together and the following proportionate mixture made:

100.00 parts Tennessee brown rock phosphate.
33.33 parts sulfur flour.
200.00 parts soil.

The plot excavations were filled with this mixture flush with the surrounding soil. Each plot received 0.02 pound of ferrous sulfate and 0.02 pound of aluminum sulfate for each 100 pounds of the compost mixture. In each case

TABLE 4

Influence of depth on the accumulation of available phosphoric acid: Available P_2O_5 calculated in pounds per ton of compost

PLOT NUMBER	DEPTH	AVAILABLE P_2O_5		
		At beginning	After 7 weeks	After 14 weeks
	inches	*pounds*	*pounds*	*pounds*
1	½	3.97	9.60	13.40
2	1	3.97	9.17	16.20
3	2	3.97	9.95	21.20
4	3	3.97	15.37	24.95
5	4	3.97	16.00	25.90

inoculation was made with soil known to be high in its sulfofying power. Cultivation was made easy by the flatness of the composts. This was the principal motive for making them flush with the ground. The composts in the preceding experiments were heaped up to a certain degree. The flat compost would represent the farmer's possibility of making available phosphorus by the sulfur-floats method in the open field. Under such conditions the soil could be stirred by ordinary tillage implements, and this would constitute the most economical method of securing adequate aeration. In this experiment the plots were cultivated once a week. Samples were taken at the beginning and moisture and available phosphorus determined. At the end of 7 and 14 weeks, respectively, samples were also secured and analyzed. The plan of the experiment and the data secured are recorded in table 4.

At the end of 7 weeks we note not only a decided gain of available phosphorus over the amount present at the beginning, but also observe comparatively wide differences between the composts themselves. The depths of ½, 1 and 2 inches are nearly identical, each containing between nine and ten pounds of available phosphoric acid. The depths of 3 and 4 inches are also identical but contain considerably more available phosphoric acid, the amount being 15.37 and 16.00 pounds of P_2O_5, respectively. As the months of June and July were somewhat rainy it is very probable that considerable leaching occurred in the very shallow composts. The relatively higher results obtained at the end of 14 weeks would substantiate this. Nevertheless, the depths of 3 and 4 inches still show the largest amount of available phosphoric acid. The large amounts of 24.95 and 25.90 pounds of P_2O_5 in such a short period of time are very gratifying, making it appear that this method of composting will prove most acceptable for practical use by the individual farmer.

SUMMARY

1. The results obtained by the authors in experiments carried on under field conditions show that the sulfur-floats-soil compost may be utilized in rendering available the phosphorus of floats, thus confirming earlier experiments reported from the laboratories of the New Jersey Agricultural Experiment Station.

2. Experiments are reported which were designed to determine the best form of compost from the standpoint of adequate aeration.

3. Temperature is an important factor in sulfur oxidation. The results obtained would indicate that for maximum efficiency composts should be made up in warm weather.

THE OXIDATION OF SULFUR BY MICROÖRGANISMS IN ITS RELATION TO THE AVAILABILITY OF PHOSPHATES[1]

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INTRODUCTION

It has already been shown in previous experiments carried out at the New Jersey Agricultural Experiment Station (44, 45, 46, 47,) that:

1. The oxidation of sulfur in sand and soils was found to render soluble in neutral ammonium citrate and water appreciable quantities of the phosphorus of raw rock phosphate.

2. The largest amount of available phosphoric acid at the end of 30 weeks was found in composts made up with a red silt loam soil. In this case 85 per cent of the phosphorus in the floats had been rendered available.

3. In both sand and soils, the oxidation of sulfur as measured in terms of sulfates, paralleled the increase of available phosphoric acid.

4. Moisture was an influential factor in the oxidation of sulfur and the making available of raw rock phosphate.

5. Vegetation experiments showed that the sulfur-floats-soil compost could be employed to advantage as a substitute for acid phosphate.

6. Experiments carried out under field conditions corroborated the results obtained in the laboratory.

In view of the above findings, it was deemed advisable to start a number of new experiments designed to involve numerous attending factors and influences. Therefore, since the above investigations were reported there has been an accumulation of valuable data, the importance of which not only surpassed expectations but predictions as well. The great value of inoculation was established and aeration shown to be a vital factor. The purpose of this paper is to set forth the results of these investigations.

Before entering upon a discussion of the results obtained it seems desirable to refer to certain publications on sulfur oxidation, in part, reviewed in an earlier paper (47), which have a more or less direct bearing on the problem at hand. The numerous investigations which have been carried out by mixing

[1] Portion of a thesis submitted to the faculty of Rutgers College in partial fulfillment of the requirements for the degree of Doctor of Philosophy, September, 1917 (Ref. no. 53).

The writer is indebted to Dr. J. G. Lipman for originally suggesting the problem and for valuable advice rendered during the progress of the work.

floats with decaying organic matter, etc., in an endeavor to render insoluble phosphates soluble, have been reviewed elsewhere (47, 49, 50). In this place it need be stated merely that the results obtained in such experiments were unsatisfactory.

HISTORICAL

The fact that elemental sulfur is transformed to sulfates in soils was recorded in 1869 by Marés (53). In answer to the question as to what becomes of sulfur which is applied to soils, he says: "It is transformed into sulfuric acid which combines with the bases present." In order to demonstrate this he made a chemical examination of a calcareous soil which had received annual additions since 1858 of elemental sulfur and also soil from the same field which had not received any sulfur. He found large amounts of water-soluble sulfates in the former soil but none in the latter. The author emphasized the importance of having the sulfur evenly distributed in the soil, and of having it in a finely divided state, if it is to be oxidized rapidly. He also noted that the transformation of sulfur into sulfates was more rapid in soils which had been fertilized. In experiments designed to determine whether hydrogen sulfide is an intermediate product in the oxidation of sulfur, he found that such was not the case. Evidently he did not know that sulfur oxidation is largely a biological process, as he states that "the surface soil is a porous layer which has a very strong oxidizing power." He concluded that: (1) Sulfur may be transformed into sulfates by direct oxidation or else hydrogen sulfide might be an intermediate product, which would be converted into sulfates as soon as formed, or under favorable conditions it would be converted into sulfuric acid which would in turn act on any bases present in the soil; and (2) in certain soils, potassium might be brought into a soluble form by the oxidation of the sulfur.

In 1877 it was suggested by Charles F. Panknin, of Charleston, S. C., that sulfur, if mixed with ground bone or ground mineral phosphates, would be oxidized to sulfuric acid when incorporated into cultivated soil. He applied for a patent on his process in April, 1877, and succeeded in August of the same year in securing Letters Patent 193,890. In his application for the patent Panknin made the following statement:

> My invention relates to a compound consisting of a mixture of finely-powdered bone or mineral phosphates and finely-powdered sulfur, the said ingredients being mixed together and applied in a dry state, and the conversion of the insoluble phosphate of lime is effected by the sulfuric acid eliminated by the natural oxidation of the sulfur of the mixture in the soil to which it is applied.
>
> To prepare the fertilizing compound, take 95 parts of the insoluble phosphates, (either bone or mineral phosphate,) in a finely powdered state, and mix with it 5 parts of finely-powdered sulfur, both ingredients being in a dry state, though I do not confine myself to these proportions. This mixture though only mechanical in its character, when applied to the soil, becomes slowly transformed, the sulfur being first converted into sulfuric acid by a natural process of oxidation, and when so converted, uniting with the insoluble phosphates to form a soluble phosphate, which is dissolved by the moisture of the soil, and acts in the same manner as phosphates made by artificial process.

In September, 1904, a patent application was submitted by William B. Chisholm of Charleston, S. C. and Patent 824,280 was secured by him on June 26, 1906. Chisholm makes the following statement:

> I find that the percentage of sulfur, by weight, to give effective results may in some instances be as low as forty pounds to the ton of the mixture—i.e., forty pounds of sulfur to nineteen hundred and sixty pounds of phosphate rock—and in other instances from sixty to one hundred pounds of sulfur to the ton of mixture. In the practical use of my invention I find that the crops produced are apparently far superior to what might reasonably be expected from the amount of available phosphate, nitrogen, and potash or other ingredients present in the final mixture. This may be due in part to the extreme fineness and intimate association of the admixed particles tending to facilitate their ready and uniform distribution through the soil, their chemical interaction under the influence of the moisture of the soil and the infiltrations of rain, and their increased solubility in said condition of impalpable powder; but I am further led to believe that the sulfur itself has an independent effect as a germicide, destroying bacterial and insect life prejudicial to the plant and either not interfering with or positively stimulating helpful bacteria.

The above citations from the Panknin and Chisholm patents bring out some very interesting facts. It is evident that Panknin recognized nearly forty years ago the value of sulfur oxidation for transforming insoluble into soluble phosphates. He did not know, of course, that the process of sulfur oxidation is largely biological in character. Neither did he recognize that the very small quantities of sulfur recommended by him, namely 1 part to 20 parts of phosphate, were entirely inadequate for accomplishing the desired reaction. The same criticism may be made of the process proposed by Chisholm. In both instances relatively small amounts of sulfur are directly applied to the soil with the expectation that the sulfuric acid ultimately produced would attack the insoluble phosphates.

In attempting to determine the value of sulfur as a fertilizer, Degrully (19) discovered in 1911, that the sulfur applied at the rate of 109 gm. per square meter was largely oxidized to sulfates.

Boullanger (7), and later Boullanger and Dugardin (10), were able to prove a favorable effect of sulfur on crop yields. Since the corresponding results in sterilized soil were not marked, they concluded that the oxidation of sulfur is brought about by bacterial activities. The results just noted were confirmed by Demolon (21, 22) in 1912 and again in 1913 (23).

In his earlier experiments he employed 5 gm. of sulfur to each 100-gm. portion of soil. Notwithstanding the relatively large application, much of the sulfur was oxidized to sulfuric acid. In other experiments he added 1 per cent elemental sulfur to a sterilized and an unsterilized soil. He found very little of the sulfur to have been oxidized in the sterilized soil at the end of two months, although a considerable amount was oxidized in the unsterilized soil. He obtained no reaction for gaseous SO_2 or H_2S in this experiment. The author thinks that sulfur added to soils may act as a source of sulfuric acid. Subsequently, Demolon (23) in another paper records experiments in which he added 1 per cent of sulfur to garden soil, kept the mixture moist and at a

temperature of 20°C. Under these conditions appreciable quantities of sulfuric acid were formed, as shown by the amounts of calcium sulfate found in the mixture at the end of 40 days. It was observed at the same time that the oxidation of the sulfur was most active in a non-sterilized soil.

In vegetation experiments it was found that when sulfur was supplied in an amount equal to that of the nitrogen, the yields were as good as those obtained from a complete fertilizer. He ascribed the beneficial action of sulfur as having been due to its influence on the activities of soil bacteria, as well as to its gradual transformation into sulfuric acid, and the reaction of the latter with basic material to form an increased supply of mineral plant-food.

The oxidation of sulfur was made the subject of study also by Brioux and Guerbet (11). They dealt particularly with the influence exerted on the oxidation of sulfur by the soil itself and by certain carbohydrates, peptone and certain other nitrogenous materials. Their investigations showed that carbohydrates had a retarding effect, while peptone accelerated it so that in 30 days 82 per cent of the 4 gm. of the flowers of sulfur added, was oxidized to sulfates. In sterile soil and in soil which had been sterilized and inoculated with soil infusion, they found 1.6 per cent and 94.2 per cent, respectively, of the 0.20 gm. of precipitated sulfur originally added, to be oxidized to sulfates in 16 days. Calcium carbonate was also found to have enhanced the rate of sulfur oxidation. Some bacteria which would oxidize sulfur were isolated, one of which would oxidize 18.6 per cent as against 3.6 per cent found in the sterile check. They concluded that the oxidation of sulfur in soils is a very complicated process which involves the activities of a number of species of bacteria. They think that perhaps the transformation is brought about in a way less direct than pure oxidation and in certain cases with the formation of hydrogen sulfide as an intermediate product.

In another experiment they found the oxidation of sulfur in soil to have exerted a solvent action on the soil minerals. By extracting the soil with distilled water they obtained a considerable increase of potassium and calcium over that obtained from the check soil. A test showed that none of the soil phosphorus had been rendered soluble.

In 1912 von Feilitzen (27) carried on some pot experiments designed to study the fertilizing action of flowers of sulfur on horse-beans and ray-grass. After harvesting the crops he tested the various soils for water-soluble sulfates and acidity. Very little difference was noted in the sulfate content of the soils, although a litmus test showed the acidity to have been greatly increased by the sulfur.

Pfeiffer and Blanck (57) during the same year found applications of sulfur at the rate of 267 and 524 pounds per acre, on field plots, to have increased the sulfate content of the soils. This increase was directly proportional to the amount of sulfur applied. The check plot was found to be neutral in reaction, while the others were strongly acid, showing that the sulfur had been oxidized to sulfuric acid.

Some interesting data concerning the influence of sulfur on soil acidity are recorded by Lint (40). In order to ascertain the rate of oxidation of sulfur in soil, 100-gm. portions of it were treated with 33 mgm. of flowers of sulfur, which is an addition equivalent to 100 pounds of sulfur per acre-foot of soil. The moisture content was 20 per cent. The acidity was found to increase up to the eighth week when all the sulfur was apparently oxidized. Experiments with a heavy clay loam and a sandy loam soil showed a much more rapid oxidation of the sulfur in the latter soil. Soils with a water content of 20 per cent when allowed to dry did not cause as rapid an oxidation as those maintained at an optimum moisture content throughout the experiment.

Somewhat later Pitz (59) found elemental sulfur to increase soil acidity, in amount proportionate to the per cent of sulfur applied, thus corroborating the work of other investigators. He also found the number of bacteria which would grow on agar plates to decrease after a certain period in a silt loam soil which had received an application of sulfur.

Experiments reported a short time later by Duley (25) confirmed the results just noted. He, however, found the sulfur to be oxidized to sulfates in both quartz sand and soil. The lime requirement of the soil which had been treated with sulfur was directly correlated with the amount of soluble sulfates formed.

Shedd (65) also studied the oxidation of sulfur in soil and sand. With soil to which sulfur was added in amounts varying from 100 pounds to 7000 pounds per acre, he found by analysis, that 60 to 80 per cent of the sulfur, regardless of the amount added, was oxidized after a period of about 4 months. Sulfur was oxidized also in the sand but not to the extent that it was in the soil.

Kappen and Quensell (35) made a study of the sulfur cycle in soils. In connection with these experiments they studied the oxidation of ground rhombic sulfur, colloidal sulfur and the so-called "Schwefel milch." The latter was prepared by mixing together 2 parts of sulfur, 1 part of calcium hydroxide and 13 parts of water. The solution was heated until the constitutents dissolved. After diluting and precipitating with dilute hydrochloric acid, the precipitated sulfur was filtered and washed. Two hundred milligrams of the dry sulfur thus prepared and of the ground rhombic sulfur were mixed with the different soils under investigation, incubated for 6 weeks, extracted with 10 per cent hydrochloric acid and the sulfates determined in the extract. The results which they obtained are quoted below to show the striking oxidation of the precipitated sulfur.

SOIL	SO_4 FROM 200 MGM. OF SULFUR	
	Rhombic sulfur	Precipitated sulfur
	mgm.	*mgm.*
Glass sand	3.69	44.63
Sandy soil	3.81	144.22
Loam soil	16.44	227.57
Meadow soil	10.91	284.01

Brown and Kellogg (13) report some experiments on the so-called sulfofying power of soils. They find that each soil has a definite sulfofying power. Organic matter was found to increase the amount of sulfur oxidized. Manure and green manure increased it up to a certain point. Manure was found to play an important part in the oxidation processes, and the optimum moisture condition was apparently 50 per cent of complete saturation. Other experiments which they carried out show that aeration is an important factor, as is also the absence of carbohydrates. The presence of bacteria is essential, although the production of a small amount of sulfates was brought about by chemical action.

Brown and Johnson (12) made a study of the effect of gypsum, acid phosphate, rock phosphate, alone and with gypsum, and mono-calcium phosphate on sulfur oxidation in an alkaline soil high in organic matter. They found all of the above materials to exert a stimulating action when applied in amounts based on actual field conditions. The rock phosphate, mono-calcium phosphate and gypsum exerted the greatest stimulating action. They report other experiments in which calcium and magnesium carbonates were employed. With small applications of calcium carbonate they obtained greatly increased sulfur oxidation, and even with excessive amounts a favorable effect was obtained. On the other hand, small applications of magnesium carbonate increased sulfofication, while large amounts exerted a depressing action.

Ames and Boltz (1), in their recent study of the effect of sulfur on the growth of plants, found that it produced increased yields when applied to certain crops. In order to determine whether or not this beneficial action was due to the solvent action on the mineral soil constituents of the acids produced during the sulfofying processes, they conducted a composting experiment in which sterilized and unsterilized soils, a loam and a clay soil, and pure quartz sand were used. To 500-gm. portions of the soil and sand were added 0.25 gm. of flowers of sulfur and Tennessese phosphate rock in amounts containing 0.06 gm. of phosphorus. To each mixture was added 400 cc. of distilled water. After two months they found but a slight increase in the availability of the phosphorus, as measured by 0.2 per cent hydrochloric acid. The acidity of the mixture had also increased but little.

Among the more recent investigations which have been made relating to sulfur oxidation, should be mentioned those of Ames and Richmond, and Brown and Warner.

Ames and Richmond (2) report experiments in which the effect of sulfur, gypsum, and acid phosphate on the changes in nitrogen and phosphorus of horse manure were studied. They found all of these materials to be very effective in reducing the loss of dry matter in manure; sulfur flowers being the most effective. The materials also reduced the loss of nitrogen from the manure. Manure treated with acid phosphate, sulfur, and calcium phosphate lost, on an average, 3.5 per cent of its total nitrogen in 250 days, as compared with a loss of 10.5 per cent from untreated manure. A study of the hydrogen

sulfide evolved from the different manure mixtures showed the gas to be given off in largest amounts from the ones treated; the largest being evolved from those treated with sulfur.

The acidity of water extracts from the mixtures after fermentation showed that the sulfur-treated manure had increased in acidity, while the untreated manure had become alkaline in reaction.

The manure treated with flowers of sulfur produced water-soluble sulfates equivalent to 23.4 gm. of sulfuric acid, as compared with a loss of about 4 gm. of sulfuric acid from the untreated manure.

Brown and Warner (14) report experiments which confirm the results reported previously by the writer and also those reported in this paper. They made up mixtures of sulfur and floats, with compost, horse manure or cow manure. These mixtures were allowed to ferment for varying lengths of time and there was found to be an enormous increase in the production of available phosphorus which continued up to fifteen weeks. The increase in available phosphorus was greater where the sulfur and floats were intimately mixed with the manure than where they were added to the manure in layers. All of the manures and the soil tested contained efficient sulfofying organisms. The authors think that "further tests of this method of producing available phosphorus may prove it to be of considerable practical value."

In view of the fact that considerable work has been carried on in which sulfur has been used in agricultural practice, the author thought that it would be advisable to give a brief résumé of at least some of the more important results obtained from the use of sulfur as a fertilizer, in both this country and in Europe; especially so since a majority of the experiments have a direct relation to the question of sulfur oxidation. From a study of them we are able to obtain some indication as to the conditions, both physical and chemical, under which the most efficient oxidation of sulfur can be obtained in soil.

SULFUR IN ITS RELATION TO PLANT GROWTH

Up to the present time elementary sulfur or its compounds have not been generally considered essential as a constituent of commercial fertilizers. Hence their use as a fertilizer has been confined almost entirely to experiments. In recent years investigations on the effect of sulfur on plant growth have been quite common. Numerous reports have been made public from time to time. Possibly this stimulus to sulfur research has been due to the conflicting nature of these reports; for some investigators record a beneficial effect of sulfur on the growth of plants, others, a detrimental effect, and still others, no effect whatever. Even more conflicting are the explanations offered as to the action of sulfur in soils.

However, it is generally agreed that plants contain, and require, sulfur, especially those rich in albuminoids. Even with the imperfect analyses of former years it was found that plants such as cabbage and alfalfa had a high

sulfur content. By an improved method of analysis Hart and Peterson (31) succeeded in demonstrating a still higher sulfur content in these and other crops. Among others Shedd (65) and Peterson (55, 56) report considerable amounts of sulfur in plants. Hart and Peterson (31) also showed that: "Normal soils are relatively poor in sulfur trioxide. An acre foot will contain from 1000 to 3000 pounds of sulfur trioxide. Soils cropped from fifty to sixty years and either unmanured or receiving but slight applications during that period have lost on the average 40 per cent of the sulfur trioxide originally present, as determined by comparison with virgin soils." These results were later confirmed by Shedd (64) who made a study of the sulfur content of Kentucky soils. Also Kossovich (36) concluded that an impoverishment of the soil as to sulfates is possible and that sulfate must be added in fertilizers. Subsequent work of Robinson (62) showed that most of the important American soils were low in sulfur. Thus an appreciable presence of sulfur in plants with a possible appreciable absence of it in soils would seem to justify the investigations that have been and are being made.

Most of these investigations indicate beneficial effects from the use of sulfates. The methods used and plants under trial have varied. Among the different sulfur compounds the sulfates were the first observed to enhance crop yields. Bogdanov (5) in 1899 found a response to fertilizers containing sulfates. In 1905 Dymond, Hughes and Jupe (26) demonstrated the value of sulfates, particularly with crops containing a high proportion of albuminoids. Lierke (38) reports that fruits responded to fertilizers containing considerable amounts of sulfates. Reimer (61), Hart and Tottingham (32) and many others also record beneficial results from the use of sulfates.

Other compounds of sulfur have also been tried. Pyrites at the rate of 89.2 to 178.5 pounds per acre were found by Vermorel and Danthony (73) to augment the yields of wheat and beans 30 to 60 per cent when used in combination with organic matter. Galtsev and Yakushkin (29) likewise found pyrites to exert a beneficial effect. Sulfuric acid tested by Rabaté (60) and by Demolon (23), sulfurous acid and carbon bisulfid tried by Demolon (23) and various sulfites employed by others were found to exert a fertilizing effect upon plants.

Quite recently there has developed a wide interest in elementary sulfur, although its fertilizing effect on vines was noted over half a century ago. In 1858 Marés (42) discussing some of his own observations, said: "On peut considerer le soufre, pour les vignes, comme un engrais, on plutot comme un amendement d'un ordre particulier."

From time to time the belief had also been expressed that sulfur where used for potato diseases was not only effective in reducing the disease but also in increasing the yield. In 1911 Chancrin and Desriot (16), using 223 to 446 pounds of sulfur per acre, noted an increase in potato and beet crops. In 1912 Chancrin and Desriot (17) again report increased yields. A continuation of this work was reported in 1913 by Desriot (24). Boullanger (8) tested flowers of sulfur in comparison with various sulfates on carrots, beans,

celery, spinach, lettuce, sorrel, chicory, onions, and potatoes. He reports in 1912 that in most cases increased yields were obtained, but that flowers of sulfur were especially favorable. In 1913 Boullanger (9) reports experiments which show the importance of sulfur as a fertilizer. Favorable results are also reported by Vercier (71), Liechti (37), and by Demolon (21, 22). Demolon (23), continuing his work, stated a year later that "potatoes were especially aided by applications of sulfur." Good results were secured by Urban (70) in the growing of sugar beets; by von Feilitzen (27) on potatoes; by Bernhard (4) on hoed crops, especially potatoes and mangolds; by Magnien (51) on turnips and beets; by Tottingham (68) on rape and radishes; by Reimer (61) on alfalfa; and by Ames and Boltz (1) on clover. Shedd (65) of the Kentucky Experiment Station, using sulfur and various compounds of sulfur as fertilizer for soybeans in pot experiments, concluded that the best results were obtained with elementary sulfur. Duley (25) found sulfur to increase markedly the yield of clover, as well as the production of nodules. He also found sulfur to increase chlorophyll production in the case of corn.

While sulfur was thus found to be beneficial the extent of the benefit very often varied. It came under the observation of some that the effect was modified by the presence or absence of organic matter. In pot experiments where sulfur was applied at the rate of 44.6 to 89.2 pounds per acre, Vermorel and Danthony (73) noted increased yields only where organic matter was added. They stated that sulfur was without effect when used on soils lacking in organic matter. Chauzit (18), in 1914, in tests with grapes, obtained the best results where sulfur was used in conjunction with a fertilizer high in organic matter. Vermorel (72) in the same year cites experiments which show that an application of manure enhanced the benefits of sulfur. Zolla (76) asserts that sulfur has a marked effect as a fertilizer when the soil is well stocked with organic matter; and that the effect of sulfur decreases as the content of organic matter is decreased. Sherbakoff (66) found the injurious after-effects of sulfur to be greatest on parts of the field poorest in humus. In accord with these findings are the observations of Sabashnikov (63) and others that sulfur does best when applied to a fertile soil.

A further modification of the effect of sulfur was found to depend upon its use in combination with fertilizers. For instance, Gianitto (30) obtained an increase in yield of potatoes when 400 pounds of sulfur per acre were applied with other fertilizer, and a decrease where the sulfur was applied alone. Chauzit (18) reports in 1914 that when 178 pounds of sulfur were used without the addition of other fertilizers, the yield of grapes was increased 2 to 20 per cent; when used with other fertilizers it was increased from 9 to 35 per cent. Similar results were noted by McConnel (48), Janicaud (34), and others.

At times the extent of the benefit from sulfur has been so small as to be negligible. In fact, its application has frequently been described as producing no results whatever. Vermorel and Danthony (73) claimed that sulfur showed no effect when used on soils lacking in organic matter and receiving

nitrogen in the form of nitrate of soda. Shedd (65) announces that in some of his experiments there was no effect produced by certain forms of sulfur. Tritschler (69) obtained but slightly increased yields of mangelwurzels. Voelcker (74) found sulfur to be without effect on mustard, rape and clover. Thalau (67) and many others term their results as "inconclusive." Bosinelli (6) thinks that the commercial use of sulfur as a fertilizer is of very doubtful value. Pfeiffer and Blanck (57) found that applications of sulfur at the rate of 267 and 524 pounds per acre did not increase the yield of oats; in fact, in many cases the opposite result was obtained. Pfeiffer and Simmermacher (58) obtained the same results a year later with beets. They concluded that the general use of sulfur as a fertilizer is not justified.

Furthermore, in the experiments of some investigators sulfur has not only failed to show a benefit, but has passed from a neutral into an injurious substance in its effect upon plant growth.

Demolon (23) in 1913 noted that cereals were injured by applications of sulfur on light soils poor in calcium carbonate. Gianitto (30), it will be recalled, noted a decreased yield of potatoes where sulfur was used alone at the rate of 400 pounds per acre. Deleterious results were likewise obtained by Janicaud (34) whenever sulfur was used alone. Although Hart and Tottingham (32) secured good results with sulfate on red clover, they found elemental sulfur to exert a harmful effect, this being attributed to incomplete oxidation of the sulfur, or to acidity resulting from the formation of sulfuric acid.

Usually the harmful effects of sulfur have been noted on the subsequent crops. It was reported by Wheeler, Hartwell and Moore (75) in 1899 that unless a considerable quantity of lime were added, injury befell cereals following potatoes which had received an application of sulfur for the prevention of potato scab. This injurious effect they ascribed to free acid in the soil formed from the slow oxidation of the sulfur. Regarding the effect of sulfur treatment on succeeding crops, Sherbakoff (66) states that where it was applied at the rate of 900 pounds per acre, there was a noticeably poor stand of clover, but where it was applied at this rate in combination with lime and additional fertilizer, the injurious effect was less marked. Also von Feilitzen (27) and Lint (41) report similar results. Lint states that in two instances where sulfur was applied in the row to potatoes, hay failed to grow in the following year—thus showing prominent bare streaks across the field where the rows had been. The evidence of this injury slowly disappeared, and by the end of summer no traces of injury remained. A year later Lint (42) reports that no appreciable injury was noticed on a succeeding crop of alfalfa where lime was applied to the soil.

Since it appears that sulfur and its compounds usually exert an influence upon the higher plants it would be natural to expect an influence, also, upon the lower forms of plant life—the microörganisms of the soil. R. Lieske (39) observes that in certain cases bacteria cause denitrification as well as sulfur oxidation. Brioux and Guerbet (11) infer that sulfur incites a complicated

bacterial process to effect its oxidation. That sulfur exerts a stimulating action on the bacteria which break down the nitrogenous matter of the soil to ammonia is the belief of some scientists; among whom are Boullanger and Dugardin (10), also Bosinelli (6). Sulfates, however were found by Fred and Hart (28) to produce no effect upon the rate of ammonification or the destruction of organic matter in the soil, and to cause no appreciable increase in the number of bacteria which would grow on agar plates. Pitz (59) obtained the same results. In the use of elemental sulfur Pitz records a decrease in the total number of bacteria but an increase in ammonification, accompanied by a parallel decrease in nitrate formation. Although there has been considerable work done by other investigators the relations in question are not well understood—beyond that the action on the soil flora, brought about by sulfur, is certainly a factor to be considered in any theory as to the effect of sulfur upon higher plants.

The rôle that sulfur plays in the soil—the theory of its action—is an undecided question. Various views have been expressed. It has been suggested by Bernhard (3), Chancrin and Desriot (16), Boullanger (8) and Heinze (33), that it functions as a disinfectant or partial sterilizer. To quote the opinion of Chancrin and Desriot: "The beneficial effects of sulfur may be due to action similar to that of partial sterilization by heat, carbon-bisulfid, toluene, etc." The belief that it has a physical effect upon the soil is maintained by Bernhard (3) and Degrully (20). A catalytic function is ascribed to it by Cettolini (15) and Boullanger (8) and others. Still other theories have been advanced by Miége (54) and many others. Worthy of mention, perhaps, is that of Demolon (23) who attributes the fertilizing effect of sulfur to its progressive transformation into sulfuric acid, thereby supplying sulfur to the plant and rendering soluble other mineral constituents of the soil. Still more worthy of mention is the suggestion recently made by Lipman (43), that the change of sulfur to sulfuric acid in the soil may be employed for converting sodium carbonate into sodium sulfate and the making productive the barren alkali areas.

In a brief review of the numerous papers on the relation of sulfur to plants it is quite necessary to omit details, or even to avoid mention of some experiments. Nevertheless, the striking importance of the subject cannot be ignored. The sulfur requirements of plants, the possible absence of it in soils and the mode or material for its application as a fertilizer are important questions to consider. Of no less interest to the scientific man are its influence on plant growth, upon microörganisms, and the theories of its action. It is safe to assume that the future will not lack in enlightening investigations upon sulfur in its relation to plant growth.

SERIES 1. FURTHER STUDIES ON ECONOMICAL SULFUR-FLOATS COMBINATIONS

Since the experiments already reported (47) dealt with relatively short incubation periods and yielded data that were somewhat conflicting, it was deemed advisable to try a similar experiment for a longer period of 24 weeks.

In this instance, however, greenhouse soil was used instead of Sassafras loam soil and all the composts were inoculated with infusion; the composts being made in the same manner. The plan of the experiment, and the ammonium-citrate-soluble P_2O_5 found in the composts at the end of 24 weeks[2] are recorded in table 1.

In considering the extent of the increase we observe that there is a consistent increase as the amount of sulfur and floats becomes larger. There is exhibited the wide variation of 107.44 mgm. with 0.6 gm. of sulfur and 2 gm. of floats to 1977.55 mgm. of available P_2O_5 with 9 gm. of sulfur and 30 gm. of floats. The point of maximum concentration was evidently not reached

TABLE 1

Influence of varying proportions of sulfur and floats on the accumulation of available phosphoric acid

LABORATORY NUMBER	ADDITIONS	AMMONIUM-CITRATE-SOLUBLE P_2O_5		
		Average	Increase over check	Increase per gram of floats
		mgm.	*mgm.*	*mgm.*
1	None	101.05		
2	15.00 gm. floats	159.25		
3	0.20 gm. floats. 0.60 gm. sulfur*	216.25	107.44	53.72
4	1.20 gm. sulfur. 4 gm. floats	498.75	382.18	95.55
5	1.80 gm. sulfur, 6 gm. floats	730.00	625.67	104.41
6	2.40 gm. sulfur, 8 gm. floats	877.50	745.51	105.69
7	3.00 gm. sulfur, 10 gm. floats	1075.00	935.15	93.52
8	3.60 gm. sulfur, 12 gm. floats	1141.25	993.64	82.80
9	3.00 gm. sulfur, 3 gm. floats	674.38	561.69	187.23
10	3.00 gm. sulfur, 6 gm. floats	891.38	767.05	127.84
11	3.00 gm. sulfur, 9 gm. floats	1061.25	925.28	102.81
12	3.00 gm. sulfur, 12 gm. floats	1270.00	1122.39	93.53
13	6.00 gm. sulfur, 20 gm. floats	2121.25	1942.60	97.13
14	7.50 gm. sulfur, 25 gm. floats	2099.60	1901.55	76.06
15	9.00 gm. sulfur, 30 gm. floats	2195.00	1977.55	65.92

* Sulfur flour was used in all the experiments reported in this paper.

although it appears to have been nearly attained in the last three composts. In the four cases where the quantity of sulfur remained the same (3 gm.) and the amount of floats increased from 3 to 12 gm., the amount of the phosphoric acid of the floats made available continued to increase, indicating that here, too, a prohibitive concentration point was not reached. From the viewpoint of increase in availability per gram of floats supplied we find, on the contrary, that the unit production diminishes beyond certain points of concentration. Starting with the 0.6 gm. of sulfur and 2 gm. of floats we secure a gradual increase up to 105.69 mgm. with 2.4 gm. of sulfur and 8 gm. of floats, beyond

[2] The methods of analysis used are reported in a previous paper (47).

which point a decrease results. Starting again with the 3–3 sulfur-floats mixture, we obtain the highest production of unit available P_2O_5, namely, 187.23 mgm., which decreases as the amount of floats increases, until with 12 gm. of floats we have but 93.53 mg. of available P_2O_5 per gram of floats. To render less bewildering any selection of the combination which is the most efficient it probably would be helpful to place a cash valuation upon the P_2O_5 made available. This might best be done by a calculation on the basis of a ton of compost; valuing the sulfur, let us say, at 1 cent per pound and the floats at $\frac{1}{2}$ cent per pound. The net amount of available P_2O_5 produced per ton of compost, and the cost per pound of this P_2O_5 which has been made available, are shown in table 2.

TABLE 2

Amount and cost of phosphoric acid rendered available per ton of compost

COMPOST NUMBER	AVAILABLE P_2O_5	COST PER POUND AVAILABLE P_2O_5
	pounds	*cents*
1		
2		
3	2.09	14.91
4	7.26	8.37
5	11.61	7.66
6	13.26	8.74
7	16.55	8.55
8	17.18	9.67
9	10.58	8.02
10	14.06	7.83
11	16.52	8.11
12	19.52	8.00
13	30.82	8.23
14	28.70	10.52
15	28.44	12.14

A study of tables 1 and 2 shows that no direct relation exists between the highest total or the highest unit quantity of available P_2O_5 and the actual cost per pound of the same. Our highest total production of 1077.55 mgm. of available P_2O_5 cost at the rate of 12.15 cents per pound. Our highest unit production of 187.23 mgm. shows a cost of 8.04 cents per pound. But our minimum cost was 7.68 cents, in the case of the mixture containing the 1.8 gm. of sulfur and 6 gm. of floats—a combination which gives a total amount of 625.67 mgm. of P_2O_5 and a unit amount per gram of floats of 104.41 mgm. Nevertheless, it cannot be said that this compost is the most economical, for other things must be considered. For instance where the cost is 7.68 cents per pound only 12.51 pounds of available P_2O_5 are produced. It would require three such composts of 1 ton each to furnish as much P_2O_5 as is made available in compost 13, where the cost was only 8.25 cents per pound. Undoubtedly, the margin of expense is here too small to warrant such an extensive handling.

However, just what ratio of sulfur to floats would be the most economical cannot be unqualifiedly stated, as it would depend largely upon environmental conditions.

It must be remembered that the length of incubation for this experiment was for a period of 24 weeks. The results are obviously much more satisfactory than those obtained in series 3, where a 12-week incubation period was used. Very likely a longer time than 24 weeks and a more efficient sulfofying flora would be more satisfactory, as a larger amount of available P_2O_5 would be formed, which would reduce the cost per pound.

If a conclusion were drawn from the data here presented it would be that a compost made up in the ratio of no. 13, which is at the rate of 120 pounds of sulfur and 400 pounds of floats to 1 ton of soil, would be the most economical. Whether or not further investigations will confirm this ratio remains to be seen.

SERIES 2. CONCENTRATED COMPOSTS

In placing any fertilizer upon a commercial basis, economy of production, a high grade of material and high degree of concentration are some of the essentials. It probably has been noted that one of the great disadvantages of the sulfur-floats method is the large amount of soil that has been used. Accordingly, it was thought advisable to try the effect of gradually decreasing the amount of soil until pure mixtures of sulfur and floats were reached. Four different kinds of mixtures were made in duplicate of about 100 gm. each. One of them contained 25 per cent of soil, another 10 per cent, another 2.5 per cent, while the fourth received no soil whatever. One set of the mixtures so prepared was used as a check, all moisture being withheld from them. It was also thought advisable to conduct another series, introducing some materials which might act as nutrients for the microörganisms, thereby intensifying sulfofication. Hence four more identical mixtures received additional treatments of 100 mgm. of ammonium sulfate, 50 mgm. of magnesium sulfate, and 25 mgm. of potassium chloride. With the exception of the checks the composts were inoculated with an infusion of a soil known to be high in sulfofying power, and the moisture maintained at an optimum content of 30 per cent throughout the incubation period. The composition of the composts and the amount of citrate-soluble P_2O_5 found in them at the end of 15 and 24 weeks are shown in table 3. Very gratifying indeed are the returns of this experiment, indicating as they do an intrinsic value for the method. The results obtained at the end of 15 weeks, although smaller, confirm those obtained at the end of the 24-week period. Hence a discussion of the results obtained for the longer incubation period only will be taken up.

As the sulfur and floats were in a very finely divided form, the composts were poorly aerated. As the results in series 5 show that aeration is a very important factor, a higher efficiency would probably have been obtained with the concentrated mixtures if they had been stirred at intervals. Considering

first compost 5, where we have the largest amount of soil (25 per cent), we observe an increase of 3073.33 mgm. of available P_2O_5 over the check. In no. 6, where the amount of soil is but 10 per cent, there is a decrease to 2498.00 mgm. of P_2O_5. A diminution of the soil in this instance seems to have been detrimental. In composts 7 and 8 there was still a decrease in available P_2O_5, the amounts being 2409.33 mgm. and 2369.57 mgm., respectively.

TABLE 3

Available phosphoric acid in the concentrated composts at the end of 15 and 24 weeks

LABORATORY NUMBER	TREATMENT	AMMONIUM-CITRATE-SOLUBLE P_2O_5		
		Average	Increase over check	Increase per gram of floats
	At end of 15 weeks			
		mgm.	*mgm.*	*mgm.*
1	25.0 gm. sulfur, 50 gm. floats, 25.0 gm. soil.....	535.00		
2	30.0 gm. sulfur, 60 gm. floats, 10.0 gm. soil.....	628.00		
3	32.5 gm. sulfur, 65 gm. floats, 2.5 gm. soil.....	673.83		
4	33.5 gm. sulfur, 67 gm. floats,.....	690.10		
5	25.0 gm. sulfur, 50 gm. floats, 25.0 gm. soil.....	1703.33	1168.33	23.37
6	30.0 gm. sulfur, 60 gm. floats, 10.0 gm. soil.....	1690.00	1062.00	17.70
7	32.5 gm. sulfur, 65 gm. floats, 2.5 gm. soil.....	1809.17	1135.34	17.46
8	33.5 gm. sulfur, 67 gm. floats.....	1909.50	1219.40	18.20
9	25.0 gm. sulfur, 50 gm. floats, 25.0 gm. soil.....	2081.67	1546.67	30.93
10	30.0 gm. sulfur, 60 gm. floats, 10.0 gm. soil.....	1966.00	1338.00	22.30
11	32.5 gm. sulfur, 65 gm. floats, 2.5 gm. soil.....	2112.25	1438.42	22.13
12	33.5 gm. sulfur, 67 gm. floats.....	1916.20	1226.10	18.30
	At end of 24 weeks			
1	Composts 1 to 4 as above.....			
5	25.0 gm. sulfur, 50 gm. floats, 25.0 gm. soil.....	3608.33	3073.33	61.47
6	30.0 gm. sulfur, 60 gm. floats, 10.0 gm. soil.....	3126.00	2498.00	41.53
7	32.5 gm. sulfur, 65 gm. floats, 2.5 gm. soil.....	3083.16	2409.33	37.07
8	33.5 gm. sulfur, 67 gm. floats	3059.67	2369.57	35.37
9	25.0 gm. sulfur, 50 gm. floats, 25.0 gm. soil.....	3316.66	2781.66	55.63
10	30.0 gm. sulfur, 60 gm. floats, 10.0 gm. soil.....	3136.00	2508.00	41.80
11	32.5 gm. sulfur, 65 gm. floats, 2.5 gm. soil.....	3109.17	2435.34	37.47
12	33.5 gm. sulfur, 67 gm. floats.....	3044.03	2353.93	35.13

Composts 9 to 12 each received an addition of 100 mgm. of ammonium sulfate, 50 mgm. of magnesium sulfate, and 25 mgm. of potassium chloride.

Concerning ourselves now with the efficiency of the different mixtures as measured by the amount of P_2O_5 made available per gram of floats added, we notice that the highest efficiency was exhibited in no. 5, which received an addition of 25 per cent soil, in which instance there was secured 61.47 mgm. P_2O_5. No. 6 shows but 41.63 mgm. and no. 7 shows 37.07 mgm. of P_2O_5 for each gram of floats. The difference between these is not great, implying that a 2.5 per cent is about as good as a 10 per cent soil dilution. But either of

these does not far surpass the straight sulfur-floats compost which gives an efficiency of 35.37 mgm. Hence it appears that the latter would be a more economical compost than either nos. 6 or 7. The results obtained with the sulfur-floats combination look very favorable.

Taking up now the additional feature of the experiment, involving the addition of the nutrients for the microörganisms (nos. 9 to 12), it must be concluded that these materials did not exert any effect one way or the other, as the results secured are practically the same as those secured in treatments 5 to 8. This, however, is only one associative method tried out in an endeavor to secure maximum production of citrate-soluble P_2O_5 in concentrated composts; perhaps others to follow will be still more encouraging.

In a review of the experiment as a whole the striking feature is the persistency with which the composts maintain their available P_2O_5 output as the soil content diminishes. Even in the total absence of soil the results are particularly encouraging. In regard to the question as to whether sufficient P_2O_5 is made available to warrant the employment of concentrated composts, it must be admitted that the data at hand do not justify an affirmative answer. The present data do prove, nevertheless, that there is a possibility that some chemical treatment may be worked out, in which a stimulation of the bacterial activities will be brought about, which will in turn speed up the sulfofication processes.

SERIES 3. EFFECT OF SULFUR ON DIFFERENT INSOLUBLE PHOSPHATES

Since the common phosphate rocks vary considerably in their chemical composition and in their degree of hardness, obviously there would be a difference in the effect that sulfur would have upon floats derived from them. In making a study of this matter, a blue rock phosphate, a Florida soft rock phosphate and a Tennessee brown rock phosphate were employed. Also a sample of Vivianite, which was high in P_2O_5, and chemically pure tricalcic phosphate were used. Composts were made with 100-gm. portions of greenhouse soil. The amount of total P_2O_5 added in the various materials was ascertained, and a record kept of the same. The amounts of citrate-soluble P_2O_5 in the various composts at the end of 15 and 21 weeks are recorded in table 4. Without going into too much detail we shall merely make the observation that differences, as surmised, do exist between these materials.

In the case of the three natural tricalcic phosphates where approximately the same number of milligrams of phosphoric acid were contained in each compost, the Florida soft rock phosphate is evidently the most responsive to the action of sulfur oxidation. For the 15-week incubation period it gave an increase of 1757.50 mgm. of available P_2O_5 over the check as compared with 1278.75 mgm. for the blue rock phosphate and 1141.87 mgm. for the Tennessee brown rock phosphate. From the 15th to the 21st week the amount of available P_2O_5 in the composts made with the above phosphates increased to

3220.00 mgm., 2080.00 mgm. and 2073.12 mgm., respectively. These large increases might be partially explained by the fact that the composts were thoroughly stirred at the end of the 15th week in order to aerate them.

That the Florida soft rock phosphate is naturally more soluble than the others, is indicated by the fact that the untreated rock is more soluble in ammonium citrate solution than the other phosphate rocks. This somewhat natural inherent availability might partly explain the increased action. The larger amount of available P_2O_5 in the natural rock might also act as a stimulant to the sulfofying microörganisms. The softness of the Tennessee brown rock phosphate is probably of no avail, for it contains greater amounts of iron and aluminum which would tend to draw into an insoluble form a larger amount of the phosphoric acid rendered soluble through the agency of the

TABLE 4

Influence of sulfur oxidation on the availability of the phosphorus in different insoluble phosphates

PHOSPHATES EMPLOYED AND TREATMENT	AMOUNT OF P_2O_5 ADDED	CITRATE-SOLUBLE P_2O_5			
		End of 15 weeks		End of 21 weeks	
		Average	Increase over check	Average	Increase over check
	mgm.	*mgm.*	*mgm.*	*mgm.*	*mgm.*
Blue rock phosphate	4410.00	93.75		87.50	
Blue rock phosphate, sulfur*	4410.00	1372.50	1278.75	2167.50	2080.00
Soft rock phosphate	4257.00	236.25		205.00	
Soft rock phosphate, sulfur	4257.00	1993.75	1757.50	3425.00	3220.00
Brown rock phosphate	4348.00	140.63		138.13	
Brown rock phosphate, sulfur	4348.00	1282.50	1141.87	2211.25	2073.12
Tricalcic phosphate, C. P.	6864.98	2213.75		2398.75	
Tricalcic phosphate, sulfur	6864.98	4250.00	2036.25	4890.00	2491.25
Vivianite	636.00	566.25		631.25	
Vivianite, sulfur	636.00	782.50	216.25	745.00	113.75

* 5 gm. sulfur employed in each case.

sulfur. In the case of the blue rock phosphate its disadvantage is probably its inherent hardness, or rather its obstinacy to solubility. The action of sulfur is also manifested in the case of the C. P. tricalcium phosphate. Although its fine state of division gave rise to a large solubility without the use of sulfur, nevertheless it will be noticed that sulfur oxidation was very effective, increasing the amount of available P_2O_5 by 2036.25 mgm. in 15 weeks and to 2491.25 mgm. in 24 weeks. The effect upon Vivianite was not so marked because the check itself ran high, yet where sulfur was added, 745.00 mgm. of available P_2O_5 were secured at the end of 21 weeks. This apparent discrepancy is made clear when it is remembered, that the soil contains P_2O_5 which may be rendered soluble. This experiment, taken in its entirety, is of a suggestive value for the selection of raw phosphate rocks for use in com-

posts. It also shows the relation that sulfur bears toward the well-known commercial phosphates in the production of soluble P_2O_5. Other things being equal, it would seem that Florida soft rock phosphate is preferable under the conditions of this experiment to the others, for the maximum yield of P_2O_5 in the sulfur composts.

SERIES 4. THE EFFECT OF FINENESS OF DIVISION

In the matter of making phosphorus available by the use of sulfur, many somewhat attractive ideas suggest themselves, one of which is the thought of bringing the materials together in a fine state of division. Fineness from the standpoint of solubility is essential for some fertilizing substances—limestone and rock phosphate, for instance. Might not then, an intimate mixture of sulfur and floats likewise result in an increased action toward solubility? A trial of this was made in the following manner; 100-gm. portions of greenhouse

TABLE 5

Influence of fineness of division of the composts on the availability of phosphoric acid

COMPOST NUMBER	PLAN OF EXPERIMENT	CITRATE-SOLUBLE P_2O_5 IN COMPOSTS	
		Average	Increase over check
		mgm.	*mgm.*
1	15 gm. floats	138.13	
2	15 gm. floats, 5 gm. sulfur (added separately to soil and mixed)	2211.25	2073.12
3	15 gm. floats, 5 gm. sulfur (ground together and mixed with soil)	2430.00	2291.87
4	15 gm. floats, 5 gm. sulfur (ground with soil)	1380.00	1241.87
5	15 gm. floats, 2.5 gm. sulfur (added separately to soil and mixed)	1028.75	890.62

soil being used in each instance. In one case sulfur and floats were mixed with the soil by hand. In another the sulfur and floats were mixed together and were then passed through a mill before mixing with the soil. In still another the sulfur, floats and soil were ground up together in a mill, optimum moisture added and after incubating for 21 weeks, analyses were made. The results obtained are recorded in table 5.

It appears that an intimate mixture of the sulfur and floats before they are mixed with the soil, tends toward an increase of available P_2O_5. The difference is not very large but is appreciable, namely, 2291.87 mgm. as against 2073.12 mgm. where the sulfur and floats were not mixed before adding them to the soil. However, of most striking significance is the lessened yield of available P_2O_5 where the soil, floats and sulfur were ground together—striking because it helps to confirm results obtained in the following series, on aeration of the composts. In that series the tendency was toward a decrease of citrate-soluble P_2O_5 as the aeration was decreased. In the case at hand it will be readily grasped that the grinding of the soil would destroy its granular texture,

producing a powder which when moistened would lose what little air it might contain, thus creating an unfavorable environment for microörganisms. Hence we secure the low figure of 1241.87 mgm. of P_2O_5 rendered available. The fifth item in the table, in which 2.5 gm. of sulfur were employed, solicits no comment other than that this amount of sulfur is not sufficient to act on 15 gm. of floats. The main issue of the experiment, i.e., the test of fineness, would seem to be quite satisfactorily settled. We are forced to conclude that fineness or intimate contact will increase the availability of the phosphorus provided that the texture of the compost is not reduced to a fineness which will prohibit biological activities through lack of proper aeration.

SERIES 5. THE EFFECT OF AERATION AND INOCULATION

In experiments previously reported (47), the red silt loam soil, high in organic matter, was found to be more effective than the Sassafras soil, low in organic matter, as a medium for the production of available phosphorus from a sulfur-floats mixture. To assume definitely that this effectiveness was due to the presence of organic matter would be precipitate and possibly incorrect. Other factors might be involved, not the least of which would be the matter of inoculation. Soils might differ widely in regard to the presence of sulfofying microörganisms, the absence of which might prevent sulfofication regardless of the organic content of the soil. In this experiment a soil high in organic matter was employed, in order to eliminate it as the limiting factor. This condition was secured by incorporating 10 per cent of old composted manure with a red silt loam soil. One-hundred-gram portions were taken and composted with 5 gm. of sulfur and 15 gm. of floats, and moisture added in amount equal to 50 per cent of the water-holding capacity of the compost. These composts were made to differ in the matter of inoculation.

The inoculation was accomplished at the time the moisture was applied, 10 cc. of the water being added in the form of an infusion made up from a soil known to be efficacious in sulfofying action. It will be noted from the plan of the experiment that two further differences existed between the composts: namely, depth and aeration. At the end of the 15th and the 22nd weeks, determinations of the ammonium-citrate-soluble phosphoric acid were made and the results obtained are recorded in table 6.

It will be remembered that the organic matter content of these composts was the same, therefore the results should have corresponded, if organic matter is the controlling factor. On the contrary, very large differences were obtained. In every case at the end of 15 weeks the inoculated portions were higher in available phosphorus than the uninoculated, which is also true for the 22-week period. The highest amount of available P_2O_5 found in the inoculated portions at the end of 15 weeks was 1532.03 mgm., and the lowest in the inoculated portions was 465.15 mgm. Seven weeks later the ratio was 2960.75 mgm. to 503.25 mgm. The amounts cannot be given unqualifiedly,

however, for the depths of the composts were not the same, and hence aeration plays an important part. It will be noted, other things being equal, that the inoculated soil at the depth of $\frac{1}{4}$ inch was evidently the most favorable for the production of available P_2O_5, with the 2-inch depth a close second, the amounts of available P_2O_5 at the end of 22 weeks being 1460.75 mgm. and 1233.25 mgm., respectively. A depth of 4 inches appeared to be unfavorable. That this was due to aeration is indicated by the high results obtained where the com-

TABLE 6

Influence of aeration and inoculation on the accumulation of available phosphoric acid

TREATMENT	DEPTH OF COMPOST	AMMONIUM-CITRATE SOLUBLE P_2O_5	
		Average	Increase over check
At end of 15 weeks			
	inches	*mgm.*	*mgm.*
Check (initial P_2O_5)		156.10	
Inoculated	$\frac{1}{4}$	1145.00	988.90
Uninoculated	$\frac{1}{4}$	707.75	551.65
Inoculated	4	767.50	611.40
Uninoculated	4	621.25	465.15
Inoculated	2	996.50	840.40
Uninoculated	2	575.00	418.90
Inoculated and stirred*	2	1688.13	1532.03
Uninoculated and stirred	2	716.50	560.40
At end of 22 weeks			
Check (initial P_2O_5)		158.00	
Inoculated	$\frac{1}{4}$	1618.75	1460.75
Uninoculated	$\frac{1}{4}$	772.50	614.50
Inoculated	4	836.25	678.25
Uninoculated	4	661.25	503.25
Inoculated	2	1391.25	1233.25
Uninoculated	2	752.50	594.50
Inoculated and stirred	2	3118.75	2960.75
Uninoculated and stirred	2	2923.75	2765.75

* Stirred thoroughly every two weeks throughout period of experiment.

posts spread out in a 2-inch layer were stirred every two weeks. The high results obtained in the $\frac{1}{4}$-inch inoculated composts and the higher results secured in the frequently-stirred inoculated composts suggest that the oxidation of sulfur is brought about by aerobic microörganisms, and that the effect of inoculation is increased by aeration. This point would seem to be of practical importance, inasmuch as it indicates that composts made up in a commercial way would not be successful unless they were inoculated and also frequently turned. Aeration, however, without inoculation appears ineffective, as shown

by the fact that at the expiration of 15 weeks there were only 560.40 mgm. of available P_2O_5 in the uninoculated stirred composts, showing no increase over the other inoculated soils which were not stirred. But 7 weeks later 2765.75 mgm. P_2O_5 was found, which is a large increase and the second highest amount secured in the entire experiment. This is accounted for by the fact that the spatula used in stirring the composts was not sterilized, thus bringing about the inoculation of the composts intended to be uninoculated. Hence aeration alone was probably not the cause of this particular increase.

The results obtained in this experiment would make it appear that for a maximum production of available P_2O_5 in sulfur-floats-soil composts, inoculation and plenty of aeration are unquestionably advantageous and should receive foremost consideration.

SERIES 6. A STUDY OF METHODS OF INOCULATION

In the foregoing investigations the inoculation was accomplished by means of an infusion. For small composts this method has been quite satisfactory. For large composts, however, it might not be practicable. Hence the suggestion was presented of using soil from an old sulfur-floats compost heap as an inoculant. Years ago when gunpowder was produced through the medium of the so-called nitre composts, such a scheme was in vogue. On these nitre or saltpeter "plantations," whenever a new compost was started, it was the practice to incorporate with it a certain amount of inoculant secured from a compost that was known to have been productive. The success of this method at that time would indicate the success of such a method in the instance at hand. To give the matter a fair test, a soil was secured which had been composted with 5 per cent of sulfur for 15 weeks. And from this soil, composts made up with a red silt loam soil were inoculated with varying amounts of 1, 4, 7 and 10 per cent. The infusions also were made from the same soil, and each quantity of infusion added to the composts represented 5 gm. of soil. The amounts of citrate-soluble P_2O_5 obtained at the end of 15 and 24 weeks are recorded in table 7.

It will be noted in studying the results obtained at the end of the 15-week period that the increases of available P_2O_5 over the checks were much the same, the 7-gm. inoculations yielding the most, where both 2.5 and 5 gm. of sulfur were employed. The amounts 659.77 mgm. and 752.69 mgm. of P_2O_5, respectively, were not as great as that secured (849.33 mgm. P_2O_5) where the inoculation was made by infusion. Evidently, the infusion was more effective, possibly because it had a more distributive effect, i.e., penetrating to all parts of the compost. At the end of 24 weeks we find the composts inoculated with infusion still in the lead, with 1872.75 mgm. of available P_2O_5. But the composts inoculated with the soil were not far behind, as we obtained 1732.74 mgm. of P_2O_5 when 10 per cent of inoculant was employed, with 5 gm. of sulfur. Where 2.5 gm. of sulfur was used the total amounts of available P_2O_5 were

lower in every case than with 5 gm. of sulfur, the highest being 1501.05 mgm. with the 4 per cent inoculant. The inoculated composts were higher in every case than the uninoculated composts. Thus we see that the use of soil from old composts bids fair as a competitor for inoculation by infusion. While,

TABLE 7

Influence of different methods of inoculation on the availability of the phosphoric acid in the composts

LABORATORY NO.	TREATMENT	AT END OF 15 WEEKS		AT END OF 24 WEEKS	
		Average citrate-soluble P_2O_5	Increase over check P_2O_5	Average citrate-soluble P_2O_5	Increase over check P_2O_5
		mgm.	*mgm.*	*mgm.*	*mgm.*
1	1 per cent composted soil, 2.5 gm. sulfur	82.43		82.39	
2	14 per cent composted soil, 2.5 gm. sulfur	82.86		78.68	
3	7 per cent composted soil, 2.5 gm. sulfur	82.71		86.11	
4	0 per cent composted soil. 2.5 gm. sulfur	81.07		81.77	
5	1 per cent composted soil. 2.5 gm. sulfur, 15 gm. floats	669.06	529.67	1536.36	1395.73
6	4 per cent composted soil. 2.5 gm. sulfur, 15 gm. floats	701.28	561.89	1641.68	1501.05
7	7 per cent composted soil, 2.5 gm. sulfur, 15 gm. floats	799.16	659.77	1583.25	1442.62
8	10 per cent composted soil, 2.5 gm. sulfur, 15 gm. floats	703.76	564.37	1395.11	1254.48
9	1 per cent composted soil, 5 gm. sulfur	83.66		86.11	
10	4 per cent composted soil, 5 gm. sulfur	84.26		81.78	
11	7 per cent composted soil, 5 gm. sulfur	83.37		83.02	
12	10 per cent composted soil. 5 gm. sulfur	82.81		83.63	
13	1 per cent composted soil, 5 gm. sulfur, 15 gm. floats	882.17	742.78	1901.37	1760.74
14	4 per cent composted soil, 5 gm. sulfur, 15 gm. floats	884.65	745.24	1856.03	1715.40
15	7 per cent composted soil, 5 gm. sulfur, 15 gm. floats	892.08	752.69	1838.68	1698.05
16	10 per cent composted soil, 5 gm. sulfur, 15 gm. floats	835.09	695.70	1873.37	1732.74
17	No composted soil, 5 gm. sulfur	83.66		81.78	
18	No composted soil, 5 gm. sulfur, 15 gm. floats	728.53	589.14	1569.82	1429.19
19	No composted soil, 5 gm. sulfur, inoculated with infusion	83.77		83.02	
20	No composted soil, 5 gm. sulfur, 15 gm. floats, inoculated with infusion	988.72	849.33	2013.38	1872.75
21	No composted soil, 15 gm. floats	139.39		140.63	

of course it was evident that such should have been the case, judging from precedents and general principles, yet in the launching of a new enterprise like this it is usually imperative to secure permanent records of many apparently obvious reactions.

SERIES 7. INFLUENCE OF SOLUBLE PHOSPHORIC ACID

In previous experiments (47) it was shown that the sulfur was oxidized more rapidly when floats were present than in the sand alone. It was thought that the microörganisms responsible for the oxidation of the sulfur require more or less phosphorus for their life activities and hence were stimulated in their ability to oxidize sulfur. The purpose of this experiment, therefore, is to study the effect of small additions of soluble P_2O_5 on sulfur oxidation, with consequent action on the insoluble rock phosphate. Consequently, composts were made with 100-gm. quantities of greenhouse soil, 5-gm. quantities of

TABLE 8

Influence of soluble phosphoric acid on the accumulation of available phosphoric acid

TREATMENT	AVAILABLE P_2O_5	
	Average	Increase over check
End of 6 weeks		
	mgm.	*mgm.*
Floats	200.00	
Floats, 0.5 gm. acid phosphate	232.79	
Floats, sulfur	1871.89	1671.89
Floats, sulfur, 0.5 gm. acid phosphate	1853.84	1621.05
End of 12 weeks		
Floats	198.40	
Floats, 0.5 gm. acid phosphate	242.81	
Floats, sulfur	2962.29	2763.89
Floats, sulfur, 0.5 gm. acid phosphate	3132.74	2889.93
End of 18 weeks		
Floats	204.10	
Floats, 0.5 gm. acid phosphate	248.75	
Floats, sulfur	3375.00	3170.90
Floats, sulfur, 0.5 gm. acid phosphate	3538.30	3289.50

sulfur flour and 15-gm. quantities of Florida soft rock phosphate which contained 28.38 per cent total P_2O_5. All the composts were inoculated with 20 cc. of a 10 per cent infusion of an old compost; 33 cc. of water was added at the beginning of the experiment, and additional quantities of 2 cc. were added at end of every 5 weeks. The composts were stirred every two weeks. The results obtained are recorded in table 8.

There is very little difference between the available P_2O_5 content of the composts receiving no addition of available P_2O_5 (except that originally present in the floats) and that of the composts receiving 0.5 gm. of acid phosphate at the end of either the 6th, 12th or 18th week. For instance, at the end of 18 weeks with no available P_2O_5 added at the beginning we obtain an increase over the

check of 3170.90 mgm. of available P_2O_5; and with an addition of 0.5 gm. of acid phosphate we obtain 3289.50 mgm. of available P_2O_5. This increase is within the limits of experimental error, and hence one would not be justified in concluding that the soluble P_2O_5 has exerted an influence one way or the other.

TABLE 9

The influence of various salts on the availability of phosphoric acid of floats using sand as a medium

TREATMENT	AMMONIUM-CITRATE-SOLUBLE P_2O_5	
	Average	Increase over check
At end of 15 weeks		
	mgm.	*mgm.*
Sulfur	17.23	
Floats	131.55	
Sulfur, floats	591.08	459.53
Sulfur, floats, 0.20 gm. $(NH_4)_2SO_4$	616.60	485.05
Sulfur, floats, 0.50 gm. $(NH_4)_2SO_4$	751.75	620.20
Sulfur, floats, 0.20 gm. $NaNO_3$	195.55	64.00
Sulfur, floats, 0.50 gm. $NaNO_3$	166.70	35.15
Sulfur, floats, 0.20 gm. $MgSO_4$	613.38	481.83
Sulfur, floats, 0.50 gm. $MgSO_4$	638.13	506.58
Sulfur, floats, 0.20 gm. $MnSO_4$	345.56	214.01
Sulfur, floats, 0.20 gm. $MgSO_4$, 0.20 gm. $(NH_4)_2SO_4$	637.70	506.15
Sulfur, floats, 0.50 gm. $MgSO_4$, 0.50 gm. $(NH_4)_2SO_4$	699.63	568.08
At end of 22 weeks		
Sulfur	17.10	
Floats	130.75	
Sulfur, floats	771.25	640.50
Sulfur, floats, 0.20 gm. $(NH_4)_2SO_4$	783.75	653.00
Sulfur, floats, 0.50 gm. $(NH_4)_2SO_4$	855.00	724.25
Sulfur, floats, 0.20 gm. $NaNO_3$	197.75	67.00
Sulfur, floats, 0.50 gm. $NaNO_3$s	172.50	41.75
Sulfur, floats, 0.20 gm. $MgSO_4$	875.00	744.25
Sulfur, floats, 0.50 gm. $MgSO_4$	830.00	699.25
Sulfur, floats, 0.20 gm. $MnSO_4$	550.00	419.25
Sulfur, floats, 0.20 gm. $MgSO_4$, 0.20 gm. $(NH_4)_2SO_4$	808.75	678.00
Sulfur, floats, 0.50 gm. $MgSO_4$, 0.50 gm. $(NH_4)_2SO_4$	823.50	692.75

SERIES 8. EXPERIMENTS WITH POSSIBLE CATALYTIC AGENTS

Before the establishment of any new method it is often wise to subject it to many apparently irrelevant tests. Hence, while the sulfur-floats-soil composts have been shown to produce results, yet there might be a possibility that certain substances might accelerate the action, so that the floats could be rendered available in a shorter period of time. To this end experiments

were conducted in which white sand and a red silt loam soil were used as media. In the case of the white sand, composts containing 100 gm. of sand, 4 gm. of sulfur, and 15 gm. of floats were made, and 18 cc. of water added per compost—10 cc. of which was in the form of infusion, made from a soil which had been composted with sulfur several weeks. The miscellaneous materials introduced, and the results obtained at the end of 15 and 22 weeks are recorded in table 9.

It will be noted that sulfur and floats alone yielded an increase of 459.53 mgm. of available P_2O_5 at the end of 15 weeks and 640.50 mgm. at the end of 22 weeks. The 0.5-gm. portions of ammonium sulfate were slightly better, yielding 620.20 mgm. and 724.25 mgm. of available P_2O_5, respectively. The largest increase of available P_2O_5—744.25 mgm.—was obtained with the 0.2 gm. of magnesium sulfate at the end of 22 weeks. Manganese sulfate seemed to retard the sulfofication processes. On the whole, the results were much

TABLE 10

The effect of chemical treatment on the sulfur oxidized in the sand

TREATMENT	SULFUR OXIDIZED			
	Sulfur	Sulfur	Average sulfur	Increase over check
	mgm.	*mgm.*	*mgm.*	*mgm.*
Check (soil, sulfur, floats), at beginning	66.80	69.20	68.00	
Nitrogenous salts added				
None	825.55	828.00	826.78	758.78
0.20 gm. $(NH_4)_2SO_4$	798.00	801.40	799.70	691.26*
0.50 gm. $(NH_4)_2SO_4$	922.48	940.50	931.49	762.49*
0.20 gm. $NaNO_3$	192.00	191.48	191.74	123.74
0.50 gm. $NaNO_3$	123.20	125.10	124.15	54.15

* The amount of sulfur contained in the ammonium sulfate subtracted.

the same, and of no special importance except in the case where sodium nitrate was employed. Here even at the end of 22 weeks there was an increase over the check of only 67 mgm. of available P_2O_5 with 0.2 gm. of sodium nitrate, and 41.75 mgm. with the 0.5-gm. additions of sodium nitrate. This is very striking and should elicit more than passing comment, as it has been observed in the field by Vermorel and Danthony (73) and Lint (41) that sulfur did not produce beneficial results when sodium nitrate was added in connection with it as a fertilizer. From the amounts of available P_2O_5 obtained in this series it appears that sodium nitrate had a prohibitive effect upon sulfofication and that this might explain the results obtained by the above investigators. Consequently, the amount of sulfur oxidized was determined in the composts receiving no special treatment, sodium nitrate, and ammonium sulfate. The results are recorded in table 10. The data show conclusively that the addition of sodium nitrate to the composts of sand, sulfur and floats had a retarding effect upon the sulfofication processes. Of the

composts analyzed those receiving the 0.5-gm. additions of ammonium sulfate show the largest amount of sulfur oxidized, namely, 762.49 mgm. On the other hand, in those receiving additions of 0.20 gm. and 0.50 gm. of sodium nitrate, the amounts of sulfur oxidized were only 123.74 mgm. and 54.15 mgm., respectively. Whether the nitrate has an injurious effect upon the sulfofying microörganisms, whether it brings about conditions in which other organisms are able to thrive and hence crowd the sulfofiers out, or whether the latter organisms are able to utilize the sodium nitrate to the exclusion of the sulfur, are problems for further investigations.

Where the red silt loam was used as a medium the experiment differed only in the following respects: (a) 22 cc. of water was added to the composts, with

TABLE 11

Influence of various materials on the availability of phosphoric acid of floats when soil is used as a medium

TREATMENT	CITRATE-SOLUBLE P_2O_5			
	At end of 15 weeks		At end of 22 weeks	
	Average	Increase over check	Average	Increase over check
	mgm.	*mgm.*	*mgm.*	*mgm.*
Sulfur	46.53		51.25	
Floats	145.40		147.75	
Sulfur, floats	1069.03	923.63	1580.00	1432.25
Sulfur, floats, 0.20 gm. $(NH_4)_2SO_4$	921.03	775.63	1405.25	1257.50
Sulfur, floats, 0.50 gm. $(NH_4)_2SO_4$	994.85	849.45	1167.75	1020.00
Sulfur, floats, 0.20 gm. Mn SO_4	1161.25	1015.85	1547.50	1399.75
Sulfur, floats, 0.50 gm. Mn SO_4	967.18	821.78	1545.00	1397.25
Sulfur, floats, 1.00 gm. peat	1128.62	983.22	1447.50	1299.75
Sulfur, floats, 1.00 gm. peat, 0.20 gm. $MnSO_4$	1095.99	950.59	1337.75	1189.00
Sulfur, floats, 5.00 gm. peat, 0.20 gm. $MnSO_4$	1158.62	1013.22	1422.50	1274.75
Sulfur, floats, 2.00 gm. horse manure	1181.00	1035.60	1230.25	1082.50
Sulfur, floats, 5.00 gm. horse manure	1334.81	1189.41	1375.00	1227.25
Sulfur, floats, 2.00 gm. composted manure	818.87	673.47	869.25	721.50
Sulfur, floats, 5.00 gm. composted manure	953.55	808.15	1180.00	1032.25
Sulfur, floats, uninoculated	761.53	616.13	945.00	797.25

1 cc. additional for each gram of organic matter added; (b) further miscellaneous substances were employed; and (c) two of the composts were not inoculated. The results tabulated in table 11 suggest that organic substances such as peat and manure are undesirable for the most efficient production of available P_2O_5 in the composts made up with this soil as a medium. Manganese sulfate made a good showing for the 22-week period, but was not quite equal to the straight sulfur-floats-soil mixture. It is perhaps needless to give this experiment much discussion, as it is largely a verification of the preceding one. Attention might be called, however, to the low results secured where no inoculation was given.

The low results obtained in one instance where composted manure was used are not easily explained.

While the amount of citrate-soluble P_2O_5 formed in the red silt loam (table 11) is higher than in the sand, the indications of the point in question do not essentially differ. In fact, one experiment is a confirmation of the other. With the evidence at hand, it would appear that a straight sulfur-floats-soil compost is more effective than one to which the above substances have been added; for the materials here employed have not shown any markedly helpful or catalytic action beyond the limit of experimental error. On the contrary, one of them, nitrate of soda, has shown a strong depressing effect. This discovery of the action of nitrate of soda on sulfur oxidation will probably prove of commercial importance both in composting and where sulfur is applied to soil in the field.

SERIES 9. FURTHER STUDIES WITH POSSIBLE CATALYTIC AGENTS

As the foreign materials introduced into the sulfur-floats composts in the preceding experiments did not exert a stimulating action, this experiment was designed to study the effect of additional materials such as charcoal, ferrous sulfate, aluminum sulfate, etc. In this experiment composts were made up with 100 gm. of Sassafras loam soil, 5 gm. of sulfur and 15 gm. of Tennessee raw rock phosphate. Thirty cubic centimeters of water were added to each compost, 10 cc. of which were soil infusion.

The inoculation in this experiment differed, however, from that used in the previous experiment. The infusion in this case was from a soil which had been inoculated 8 weeks previously with soil from one of the very efficient composts made up with the greenhouse soil. To this soil had also been added small quantities of sulfur from time to time in order to ascertain if an efficient inoculating material could not be secured. In order to supply adequate aeration the composts were stirred every two weeks. The various materials added and the results obtained at the end of 9 and 15 weeks are recorded in table 12.

Taking into consideration the results obtained at the end of the 9-week period, it will be noted that the composts made up with sulfur-floats and soil alone yielded 1382.20 mgm. of available P_2O_5. This shows very clearly the high efficiency of the inoculating material because in another experiment where the same soil and same materials were used we obtained an increase of only 639.18 mgm. of available P_2O_5 in 30 weeks, or, in other words, we obtained 743.02 mgm. more available P_2O_5 in this experiment with no other treatment than inoculation in a period of 9 weeks than was obtained previously (47) in 30 weeks in the uninoculated soil.

Comparing the results obtained where the various materials were introduced with the untreated composts, it will be noted that three of the treatments have brought about large increases in the available P_2O_5. The addition of 0.02-

and 0.05-gm. quantities of ferrous and aluminum sulfate in combination, and a combination of 0.05-gm. quantities of ferrous sulfate, aluminum sulfate and sodium silicate, brought about considerable increases in available P_2O_5

TABLE 12

Influence of ferrous sulfate, aluminum sulfate and other materials on the accumulation of available phosphoric acid

SPECIAL TREATMENT	AMMONIUM-CITRATE-SOLUBLE PHOSPHORIC ACID					
	End of 9 weeks			End of 15 weeks		
	Average	*Increase over check	*Increase due to treatment	Average	*Increase over check	*Increase due to treatment
	mgm.	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*
Check (floats†)	111.25			120.50		
Check (floats, 0.02gm. $FeSO_4.7H_2O$)	138.75			126.25		
Check (floats, 0.02 gm. $Al(SO_4)_3.18H_2O$)	126.25			117.50		
Check (floats, 0.02 gm. $FeSO_4.7H_2O$, 0.02 gm. $Al_2(SO_4)_3.18H_2O$)	117.50			124.75		
None (floats and sulfur only)	1505.63	1382.20		2945.00	2822.75	
0.20 gm. calcium sulfate	1470.00	1346.57		2935.00	2812.75	
0.50 gm. calcium sulfate	1373.75	1250.32		2585.00	2462.75	
0.20 gm. powdered charcoal	1688.75	1565.32	183.12	2490.00	2367.75	
0.50 gm. powdered charcoal	1742.50	1619.07	236.87	3396.25	3274.00	451.25
0.02 gm. $FeSO_4.7H_2O$, 0.02 gm. $Al_2(SO_4)_3.18H_2O$	2261.83	2138.40	756.20	4162.50	4040.25	1217.50
0.05 gm. $FeSO_4.7H_2O$, 0.05 gm. $Al_2(SO_4)_3.18H_2O$	2221.75	2098.32	716.12	3788.75	3666.50	843.75
0.02 gm. $FeSO_4.7H_2O$, 0.02 gm. $Al_2(SO_4)_3.18H_2O$, 0.02 gm. $Na_2Si_4O_9$	1541.25	1417.82	35.62	3860.00	3737.75	915.00
0.05 gm. $FeSO_4.7H_2O$, 0.05 gm. $Al_2(SO_4)_3.18H_2O$, 0.05 gm. $Na_2Si_4O_9$	2108.13	1984.70	602.50	3542.50	3420.25	597.50
0.05 gm. $Al_2(SO_4)_3.18H_2O$	1661.25	1537.82	155.62	3636.25	3514.00	691.25
0.05 gm. $FeSO_4.7H_2O$	1678.75	1555.32	173.12	3777.50	3655.25	832.50
0.05 gm. $Na_2Si_4O_9$	1683.13	1559.70	177.50	3217.50	3095.25	272.50
0.02 gm. $KMnO_4$	1417.50	1294.07		2965.00	2842.75	20.00
0.01 gm. KI	1336.25	1212.82		1975.00	1852.75	
0.05 gm. KI	169.38	45.95		187.50	65.25	
0.01 gm. $CuSO_4$	1280.00	1156.57		2276.25	2154.00	
0.05 gm. $CuSO_4$	1266.25	1142.82		2683.75	2561.50	
0.01 gm. $ZnSO_4$	1757.50	1634.07	251.87	3136.25	3014.00	191.25
0.05 gm. $ZnSO_4$	1150.00	1026.57		2395.00	2272.75	

* Average of the four checks subtracted in each case.

† Floats contained 28.92 per cent of P_2O_5.

NOTE:—Checks received no sulfur.

over that found in the untreated composts. the amounts being 756.20 mgm., 716.12 mgm. and 602.50 mgm., respectively. With 0.20- and 0.50-gm. quantities of charcoal, 0.02-gm. quantities of ferrous sulfate, aluminum sulfate and sodium silicate in combination, and 0.05-gm. quantities of the three last-named

salts alone, and with 0.01 gm. of zinc sulfate, there is, apparently, a slight stimulating effect. All the other treatments brought about decreases of varying degree, the greatest retardation being caused by the 0.05 per cent addition of potassium iodide. In this case sulfofication processes were apparently stopped altogether.

The results obtained at the end of the 15-week incubation period show practically the same relative differences as those for the 9-week period, although of course, the amounts of phosphoric acid rendered available are greater. The addition of a combination of 0.02 gm. each of ferrous and aluminum sulfates has brought about an increase of 1217.50 mgm. of citrate-soluble phosphoric acid over the untreated composts, which is the largest increase for any of the treatments. In order to check the results obtained with the ferrous and aluminum sulfates in this experiment, the following investigation was carried out.

TABLE 13

Influence of ferrous and aluminum sulfates on the accumulation of available phosphoric acid

LABORATORY NUMBER AND ADDITIONS	CITRATE-SOLUBLE P_2O_5	
	Average	Increase over check*
	mgm.	*mgm.*
1. Floats (Tennessee rock 28.92 per cent P_2O_5)	121.38	
2. Floats, 0.02 gm. ferrous sulfate	138.75	
3. Floats, 0.02 gm. aluminum sulfate	126.25	
4. Floats, 0.02 gm. aluminum sulfate, 0.02 gm. ferrous sulfate	122.63	
5. Floats, sulfur	2437.50	2310.25
6. Floats, sulfur, 0.02 gm. aluminum sulfate	2927.50	2800.25
7. Floats, sulfur, 0.02 gm. ferrous sulfate	3196.25	3069.00
8. Floats, sulfur, 0.02 gm. ferrous sulfate, 0.02 gm. aluminum sulfate	3586.25	3459.00
9. Floats, sulfur, 0.04 gm. ferrous sulfate, 0.04 gm. aluminum sulfate	3526.25	3399.00

* Obtained by subtracting average amount of available P_2O_5 in composts 1, 2, 3 and 4.

In this experiment 5-gm. portions of sulfur flour and 15-gm. portions of Tennessee raw rock phosphate were added to 100-gm. portions of Sassafras loam soil. The moisture was maintained at 33 per cent and as in the preceding experiment the composts were stirred every two weeks. The citrate-soluble P_2O_5 was determined in the various composts at the end of 15 weeks. The composition of the composts and the amount of available P_2O_5 contained in them at the end of 15 weeks are recorded in table 13.

The addition of ferrous and aluminum sulfates, both alone and in combination, have brought marked increases in the available P_2O_5 of the composts. Thus we find an increase of 2310.25 mgm. of P_2O_5 in compost 5, which had received no addition of ferrous or aluminum sulfate, while in composts 6 and 7, to which had been added 0.02 gm. of aluminum sulfate and 0.02 gm. of ferrous sulfate, respectively, the amount of available P_2O_5 was increased, compost

6 yielding 2800.25 mgm., and compost 7, 3069.00 mgm. The largest increases, however, were brought about by the addition of the salts in combination. In this case we obtain an increase of 3459.00 mgm. and 3399.00 mgm. of available P_2O_5 when the salts were added in combination in 0.02-gm. and 0.04-gm. quantities, respectively.

The facts noted in the preceding experiment are thus amply confirmed. That the increase in availability of the composted floats is brought about by a stimulating of the sulfofying microörganisms rather than by a chemical reaction is clearly indicated. We find in composts 1, 2, 3, and 4 the available P_2O_5 to be 121.38, 138.75, 126.25 and 122.63 mgm., respectively. There was very little difference between the amount of available P_2O_5 in compost 1, which was composed of only floats and soil, and that in composts 2, 3 and 4 which were composed of floats, soil and ferrous or aluminum sulfate, or the two sulfates in combination. This shows that the phosphorus of the floats was not rendered available by any chemical reaction between the ferrous or aluminum sulfates and the floats.

Considered as a whole, the results obtained in this experiment and in the preceding one show that in order to obtain the maximum production of available phosphoric acid in the compost heap, in the shortest period of time, 0.4 pound each of ferrous and aluminum sulfates should be added to each ton of compost.

SERIES 10. THE EFFECT OF CALCIUM CARBONATE ON THE AVAILABILITY OF PHOSPHORUS

As the process of sulfofication is as yet not entirely understood and as there are, no doubt, many attending factors which influence it, it was thought expedient to study the effect of calcium carbonate. The influence of calcium carbonate has often been suggested, and Brioux and Guerbet (11), and Brown and Kellogg (13) found it in their experience to be a stimulant to sulfofication. An experiment was accordingly planned for this purpose. Sassafras loam soil in 100-gm. quantities was mixed with sulfur and floats and with calcium carbonate, and each compost inoculated with fresh soil infusion. For 12 weeks, optimum moisture conditions were carefully maintained. Then acidity and citrate-soluble phosphoric acid determinations were made. It will be noted that the experiment naturally divides itself into three parts. In the first, comprising composts 1 to 6, we have 0.25 gm. of calcium carbonate and 5 gm. of floats, with varying amounts of sulfur. In the second including compost 7 to 12 we have 0.5 gm. of calcium carbonate and 10 gm. of floats with varying quantities of sulfur. In the third group, composed of composts 13 to 16 there are fixed amounts of sulfur and floats with varying amounts of calcium carbonate.

The acidity and available phosphoric acid which were found at the end of the 12-week period are recorded in table 14.

Taking this experiment as a whole it cannot be considered entirely satisfactory either from the standpoint of the presence or absence of calcium carbonate. Possibly the experiment was not fair in regard to the length of time. A longer incubation period might have contributed altogether different results. And, although the treatment in regard to inoculation and moisture content, as well as to temperature and light, were apparently the same, yet possibly differences might have existed. Reviewing the results as a whole, when we consider the total increases and decreases, it is interesting to note that when calcium carbonate was added, there were five instances of increases in acidity totaling 202.00 cc. of N/50 KOH as against four decreases totaling 170.00 cc.

TABLE 14

Influence of lime on the accumulation of available phosphoric acid

LABORATORY NUMBER	TREATMENT	ACIDITY, CC. N/50 KOH	INCREASE IN ACIDITY DUE TO $CaCO_3$, CC. N/50 KOH	CITRATE-SOLUBLE P_2O_5	INCREASE IN P_2O_5 DUE TO $CaCO_3$
				mgm.	*mgm.*
1	2 gm. sulfur, 5 gm. floats	578.00		265.15	
2	2 gm. sulfur, 5 gm. floats, 0.25 gm. $CaCO_3$	800.00	22.00	273.82	8.67
3	4 gm. sulfur, 5 gm. floats	534.00		262.67	
4	4 gm. sulfur, 5 gm. floats, 0.25 gm, $CaCO_3$	596.00	62.00	158.59	− 4.08
5	5 gm. sulfur, 5 gm. floats	556.00		260.19	
6	5 gm. sulfur, 5 gm. floats, 0.25 gm. $CaCO_3$	596.00	30.00	280.01	19.82
7	2 gm. sulfur, 10 gm. floats	472.00		351.88	
8	2 gm. sulfur, 10 gm. floats, 0.50 gm. $CaCO_3$	500.00	28.00	361.79	9.91
9	4 gm. sulfur, 10 gm. floats	486.00		292.40	
10	4 gm. sulfur, 10 gm. floats, 0.50 gm. $CaCO_3$	452.00	−34.00	351.88	59.48
11	5 gm. sulfur, 10 gm. floats	578.00		433.65	
12	5 gm. sulfur, 10 gm. floats, 0.50 gm. $CaCO_3$	508.00	−70.00	361.79	−61.86
13	5 gm. sulfur, 15 gm. floats	526.00		436.87	
14	5 gm. sulfur, 15 gm. floats, 0.25 gm. $CaCO_3$	522.00	− 4.00	364.27	−72.60
15	5 gm. sulfur, 15 gm. floats, 0.50 gm. $CaCO_3$	586.00	60.00	416.30	52.03
16	5 gm. sulfur, 15 gm. floats, 1.00 gm. $CaCO_3$	524.00	−62.00	394.00	−22.30

For the phosphoric acid made available there were five cases of increases aggregating 159.84 mgm. It would appear, therefore, that in this experiment the presence of calcium carbonate exerted neither a marked beneficial nor a detrimental influence.

SERIES 11. EFFECT OF CALCIUM CARBONATE

As the results obtained in the preceding experiment were somewhat inconclusive, this experiment was planned with the idea of securing a check on the foregoing results as well as to determine the effect of larger quantities of lime as a stimulant to sulfofication processes.

One hundred-gram quantities of greenhouse soil, 5-gm. portions of sulfur

flour and 15-gm. portions of Florida soft rock phosphate were employed. Duplicate composts were made for each treatment. All the composts were inoculated with an infusion of a soil known to be very high in sulfofying power. The composts were stirred every two weeks. The plan of the experiment and the results obtained at the end of 8 and 15 weeks, respectively, are recorded in table 15.

In the composts which did not receive additions of calcium carbonate we note that from the 4354 mgm. of insoluble phosphoric acid there has been rendered available 1866.25 and 3468.75 mgm. of phosphoric acid at the end of the 8- and 15-week periods, respectively. At the end of 8 weeks the composts

TABLE 15

Influence of lime on the accumulation of available phosphoric acid

TREATMENT AND ADDITIONS	AVAILABLE P_2O_5	
	Average	Increase over check
At end of 8 weeks		
	mgm.	*mgm.*
Floats	200.00	
Floats, sulfur	2066.25	1866.25
Floats, sulfur, 0.20 gm. $CaCO_3$	1950.00	1750.00
Floats, sulfur, 0.40 gm. $CaCO_3$	2053.75	1853.75
Floats, sulfur, 0.60 gm. $CaCO_3$	2076.25	1876.25
Floats, sulfur, 0.80 gm. $CaCO_3$	2138.75	1938.75
At end of 15 weeks		
Floats	197.50	
Floats, sulfur	3666.25	3468.75
Floats, sulfur, 0.20 gm. $CaCO_3$	3305.00	3107.50
Floats, sulfur, 0.40 gm. $CaCO_3$	3442.50	3245.00
Floats, sulfur, 0.60 gm. $CaCO_3$	3551.25	3353.75
Floats, sulfur, 0.80 gm. $CaCO_3$	3355.00	3157.50

which had received 0.20-gm., 0.40-gm., 0.60-gm. and 0.80-gm. additions of calcium carbonate were found to contain practically the same amount of available phosphoric acid as the untreated, showing a slight increase where 0.80 gm. of calcium carbonate was added. At the end of 15 weeks, however, we find that the addition of calcium carbonate had brought about a decrease in available phosphoric acid in every case. The experiments thus show that lime is not a necessary adjunct to the sulfur-floats composting process.

SERIES 12. THE EFFECT OF PEAT

Among other things organic matter has in some instances suggested itself as an aid in accelerating the oxidation of sulfur in composts, thus making more of the phosphorus of floats available in a shorter period of time than in the

previous composting experiments. To give this a somewhat more thorough trial it was deemed advisable to try a partially mineralized form of organic matter in soil and sand. Since peat or its extractions are often used in biological experiments, and also in a commercial way as a medium for microorganisms, this material was selected. Composts were made up from Sassafras loam soil and white sea sand. The composts containing 4 gm. of sulfur and 15 gm. of floats were all inoculated with a soil infusion supposedly well supplied with sulfur oxidizers. Moisture conditions which were considered optimum were maintained for each treatment, 0.66 cc. of water being added extra for

TABLE 16

The available phosphoric acid in the composts as effected by peat

SOIL MEDIUM AND ADDITIONS	AT END OF 15 WEEKS	AT END OF 24 WEEKS
	Average Citrate-soluble P_2O_5	Average Citrate-soluble P_2O_5
	mgm.	*mgm.*
100 gm. soil, floats.*	152.25	138.55
95 gm. soil, 5 gm. peat	159.35	139.00
70 gm. soil, 30 gm. peat	247.85	187.80
100 gm. soil, floats, sulfur†	1616.50	3310.00
95 gm. soil, 5 gm. peat sulfur, floats	1534.29	2990.00
90 gm. soil, 10 gm. peat, sulfur, floats	1487.16	3147.50
80 gm. soil, 20 gm. peat, sulfur, floats	1128.08	2057.50
70 gm. soil, 30 gm. peat, sulfur, floats	1326.45	1801.00
No soil, 50 gm. peat, sulfur, floats	472.96	993.00
100 gm. sand, floats	146.10	125.05
95 gm. sand, 5 gm. peat	150.58	122.90
70 gm. sand, 30 gm. peat	215.36	166.88
100 gm. sand, floats, sulfur	567.74	828.50
95 gm. sand, 5 gm. peat, sulfur, floats	1083.02	1260.00
90 gm. sand, 10 gm. peat, sulfur, floats	1037.90	1327.50
80 gm. sand, 20 gm. peat, sulfur, floats	1096.40	1400.00
70 gm. sand, 30 gm. peat, sulfur, floats	1069.32	1576.25

* 15 gm. floats added in each case.

† 4 gm. sulfur added in each case.

each gram of peat. The arrangement of the series and the amounts of citrate-soluble P_2O_5 contained in them at the end of 15 and 24 weeks, respectively, are shown in table 16.

The analyses of ammonium-citrate-soluble P_2O_5 at the end of the 15- and 24-week periods show peat to have a widely different effect on the sulfofication processes in the loam soil and the sand. In the loam soil the highest results were obtained where peat was omitted at the end of both the 15- and the 24-week periods, the amounts of citrate-soluble P_2O_5 being 1616.50 mgm. and 3310.00 mgm., respectively. Where peat was used in the soil-sulfur-floats composts, there was a tendency toward a decrease of citrate-soluble P_2O_5

as the amounts of peat increased, until we get the lowest results of all where peat only was used; in this case only 993.00 mgm. of P_2O_5 from the floats was made available in 24 weeks.

In the case of the sand, just the opposite effects are noted. The lowest results occurred where no peat was added to the sulfur-floats combinations and while the production of available P_2O_5 at the end of the 15-week interval did not vary as the peat ratio varies, the tendency 9 weeks later was toward an increase of citrate-soluble P_2O_5 as the proportions of peat increased, 5 gm. of peat resulting in 1260.00 mgm., and 30 gm. of peat, 1576.25 mgm. of P_2O_5.

An important point that should be mentioned in this connection is that the increase brought about in the sea sand by the addition of peat is not an economical one, for the greatest yield of available P_2O_5 in the sand (1576.25 mgm.) is less than one-half the highest yield in the loam soil (3310.00 mgm.). Treatment for treatment, much more P_2O_5 is made available in the loam soil than in the sea sand. It appears that in the soil there are some favorable factors which do not exist in the sand, such as the ability to absorb certain toxic substances. Therefore peat, even where it did increase the quantity of available P_2O_5, was not in sufficient quantity to warrant its use. Thus this experiment strongly indicates that peat, from the commercial standpoint, at least, is not only an unnecessary but an undesirable adjunct to a sulfur-floats soil compost for the purpose of making available phosphoric acid.

SERIES 13. EFFECT OF PEPTONE ON THE AVAILABILITY OF PHOSPHORUS

In order to determine whether peptone would increase sulfur oxidation, and in this way increase the production of available phosphoric acid in the composts, 100-gm. quantities of Sassafras loam and greenhouse soil were each mixed with sulfur and floats. Both the Florida soft rock and the Tennessee brown rock phosphate were used. The ammonium-citrate-soluble phosphoric acid was determined at the end of 8 and 16 weeks, respectively. The composition of the composts and the average amount of available phosphoric acid contained in them at the end of these periods are given in table 17.

It will be seen from the results given in table 17 that a striking depression of the available phosphoric acid in the composts was caused by the presence of peptone. The addition of peptone has probably encouraged the development of certain species of bacteria which are not sulfofiers, and thus suppressed the active sulfofiers. Other reasons which could be advanced for this depression are: (a) the sulfofying microörganisms may not be able to develop where large quantities of soluble organic matter are present, being in this respect like the nitrifying bacteria; or (b) the species of microörganisms which are encouraged to develop may change the sulfur into a form other than sulfuric acid.

In order to determine what changes the sulfur in the compost had undergone, a qualitative test was made on all the composts. This test showed that

the sulfur was transformed only to sulfates in the composts in which peptone was absent. On the other hand, in the composts which had received 1 per cent of peptone, only a small amount of sulfates were present, although a

TABLE 17

Influence of peptone on the availability of floats in sulfur-floats-soil composts

COMPOSITION OF COMPOSTS	CITRATE-SOLUBLE P_2O_5 IN ENTIRE COMPOST			
	Sassafras loam		Greenhouse soil	
	Average	Increase over check	Average	Increase over check
	mgm.	*mgm.*	*mgm.*	*mgm.*
At end of 8 weeks				
15 gm. Florida soft rock phosphate	192.50		197.17	
15 gm. Florida soft rock phosphate, 1 gm. peptone	199.00		201.10	
15 gm. Tennessee brown rock phosphate,	125.75		130.00	
15 gm. Tennessee brown rock phosphate, 1 gm. peptone	135.10		136.10	
15 gm. Florida soft rock phosphate, 5 gm. sulfur	1515.57	1323.07	1983.54	1786.37
15 gm. Tennessee brown rock phosphate, 5 gm. sulfur	1490.54	1364.79	1610.00	1480.00
15 gm. Florida soft rock phosphate, 5 gm. sulfur 1 gm. peptone	220.92	21.90	206.67	5.57
15 gm. Tennessee brown rock phosphate, 5 gm. sulfate, 1 gm. peptone	149.65	14.55	125.90	−10.20
At end of 16 weeks				
15 gm. Florida soft rock phosphate	188.00		186.75	
15 gm. Florida soft rock phosphate, 1 gm. peptone	200.50		198.00	
15 gm. Tennessee brown rock phosphate	124.50		118.50	
15 gm. Tennessee brown rock phosphate, 1 gm. peptone	129.25		127.75	
15 gm. Florida soft rock phosphate, 5 gm. sulfur	3690.00	3502.00	3566.00	3379.25
15 gm. Tennessee brown rock phosphate, 5 gm. sulfur	3284.00	3159.50	3180.50	3062.00
15 gm. Florida soft rock phosphate, 5 gm. sulfur, 1 gm. peptone	218.50	18.00	178.50	−19.50
15 gm. Tennessee brown rock phosphate, 5 gm. sulfur, 1 gm. Peptone	155.25	26.00	200.00	72.25

large quantity of the sulfur had been changed into sulfites. Therefore, as no sulfites were present in the composts which had received no addition of peptone, it appears that the peptone has brought about conditions under which organisms which are able only to transform the sulfur into sulfites can develop.

SUMMARY

Under the conditions of the experiment the following conclusions appear to be justified.

1. A compost composed of 100 parts of soil, 120 parts of sulfur, and 400 parts of floats appears to be the most economical combination for the production of available phosphoric acid.

2. An appreciable amount of phosphoric acid is rendered available in composts consisting of only sulfur and floats, although the quantity is not so large as when soil is present. This indicates that a stimulation of sulfofication processes may be brought about by some treatment, and in this way enable us to obtain maximum results in composts without the aid of soil.

3. Of the natural tricalcium phosphates (Tennessee brown rock, Tennessee blue rock and Florida soft rock) Florida soft rock phosphate appears to be the most preferable for maximum yields of available phosphoric acid in composts containing sulfur.

4. Fineness or intimate contact of the constituents in composts will increase their efficiency, provided that the texture is not reduced to a fineness which will prohibit biological activities through lack of proper aeration.

5. The biological factor is influential in the oxidation of sulfur and the rendering available of phosphorus in floats. In inoculated composts, all other conditions being the same, the quantity of phosphorus rendered available in 9 weeks was double that made available in uninoculated composts in 30 weeks.

6. In making composts the question of aeration should receive foremost consideration. The results obtained in this series of investigations would make it appear that the microörganisms which oxidize sulfur are largely aerobic, and hence require an abundant supply of oxygen.

7. The presence of soluble phosphoric acid other than that contained in floats, will exert no influence on the production of available phosphorus in soil-sulfur-floats composts.

8. Ammonium sulfate, magnesium sulfate, calcium sulfate and zinc sulfate were found to exert no influence on the production of available phosphorus, while sodium nitrate, potassium iodide and copper sulfate exerted a marked depressing action. The addition of 0.2 per cent of sodium nitrate was found to inhibit sulfofication processes almost entirely.

9. Under certain conditions ferrous sulfate, aluminum sulfate, and a combination of the two salts exert a marked stimulating action on sulfur oxidation processes, when present in small amounts in a compost composed of soil, sulfur flour, and Tennessee brown rock phosphate. A mixture of the two sulfates caused the greatest stimulation. From the data which are advanced it appears that in composting under farm conditions, 0.4 pound each of aluminum and ferrous sulfates should be added to each ton of compost, in order to obtain a maximum production of available phosphorus.

10. Calcium carbonate may bring about a decrease in available phosphorus when added to a sulfur-floats-soil compost.

11. A compost is more efficient in producing available phosphorus in the absence of large amounts of organic materials, such as peat, fresh horse manure, old composted manure, and peptone. The more soluble materials will bring about the greatest decrease in efficiency.

12. Sulfofying microörganisms may develop more rapidly in the absence of soluble organic matter. They appear in this respect to be similar to the nitrifying organisms.

13. When peptone was added to a mixture of soil, sulfur and floats. sulfur was largely transformed into sulfites.

REFERENCES

(1) Ames, J. W., and Boltz, G. E. 1916 Sulfur in relation to soils and crops. Ohio Agr. Exp. Sta. Bul. 292, p. 221–256.

(2) Ames, J. W., and Richmond, T. E. 1917 Fermentation of manure treated with sulfur and sulfates; changes in nitrogen and phosphorus content. *In* Soil Sci., v. 4, no. 1, p. 79–89.

(3) Bernhard, A. D. 1910 Versuche zur Bekämpfung des Kartoffelschorfes. *In* Deut. Landw. Presse, Bd. 37, Heft 18, p. 204–205.

(4) Bernhard, A. D. 1912 Versuche über die Wirkung des Schwefels als Dung im Jahre 1911. *In* Deut. Landw. Presse, Bd. 39, Heft 23, p. 275.

(5) Bogdanov, S. 1899 Soderzhanie siery v' rasteniiakh. *In* Zhur. Russ. Fiz. Khim. Obshch., v. 31, p. 471–477.

(6) Bosinelli, C. 1915 Intorno alla azione dello zolfo libero sulla vegetazione. *In* Staz. Sper. Agr. Ital., v. 48, no. 3, p. 175–184.

(7) Boullanger, E. 1912 L'action du soufre en fleur sur la vegetation. *In* Compt. Rend. Acad. Sci. (Paris), t. 154, no. 6, p. 369–370.

(8) Boullanger, E. 1912 Études experimentales sur la engrais cataliques. *In* Ann. Sci. Agron., t. 4, ser. 1, no. 3, p. 161–180.

(9) Boullanger, E. 1913 Nouvelles experiences sur l'action fertilisante du soufre. *In* Vie Agr. et Rurale, t. 2, no. 9, p. 247–248.

(10) Boullanger, E., and Dugardin, M. 1912 Mecanisme de l'action fertilisante du soufre. *In* Compt. Rend. Acad. Sci. (Paris), t. 155, no. 4, p. 327–329.

(11) Brioux, Ch., and Guerbet, M. 1913 E'volution du soufre dans le sol, étude sur son oxydation. *In* Compt. Rend. Acad. Sci. (Paris), t. 156, no. 19, p. 1476–1479. *In* Ann. Sci. Agron., t. 4, ser. 2, no. 4, p. 385–396.

(12) Brown, P. E., and Johnson, H. W. 1916 Studies in sulfofication. *In* Soil Sci., v. 1, no. 4, p. 339–362.

(13) Brown, P. E., and Kellogg, E. H. 1914 Sulfofication in soils. Iowa Agr. Exp. Sta. Res. Bul. 18, p. 49–111.

(14) Brown, P. E., and Warner, H. W. 1917 Production of available phosphorus from rock phosphate by composting with sulfur and manure. *In* Soil Sci., v. 4, no. 4, p. 269–282.

(15) Cettolini, Sante. 1915 Experimenti di concimazione con sostanze complementari (catalitici) della cenere della vite. *In* Bol. Quind. Soc. Agr. Ital., v. 20, no. 13, p. 431–438.

(16) Chancrin, E., and Desriot, A. 1911 Action du soufre comme engrais. *In* Jour Agr. Prat., n. s., t. 21, no. 14, p. 427–429.

(17) CHANCRIN, E., AND DESRIOT, A. 1912 Influence du soufre sur la vegetation des pommes de terre et des betteraves. *In* Jour. Agr. Prat., n. s., t. 23, no. 12, p. 365–367.

(18) CHAUZIT, J. 1914 L'action fertilisante du soufre sur la vigne. *In* Rev. Vit., t. 41, no. 1052, p. 175–179.

(19) DEGRULLY, L. 1911 L'action fertilisante du soufre. *In* Prog. Agr. et Vit., t. 57, p. 321–324.

(20) DEGRULLY, L. 1912 L'action fertilisante du soufre. *In* Prog. Agr. et Vit. (Ed. l'Est-Centre), t. 33, no. 11, p. 321–324.

(21) DEMOLON, A. 1912 Sur l'action fertilisante du soufre. *In* Sta. Agron. Aisne Bul., p. 33–41.

(22) DEMOLON, A. 1912 Sur l'action fertilisante du soufre. *In* Compt. Rend. Acad. Sci. (Paris), t. 154, no. 8, p. 524–526.

(23) DEMOLON, A. 1913 Recherches sur l'action fertlisante du soufre. *In* Compt. Rend. Acad. Sci. (Paris), t. 156, no. 9, p. 725–728.

(24) DESRIOT, A. 1913 Nouvelles experiences sur l'action fertilisante du soufre. *In* Jour. Agr. Prat., n. s., t. 25, no. 12, p. 364–365.

(25) DULEY, F. L. 1916 The relation of sulfur to soil productivity. *In* Jour. Amer. Soc. Agron., v. 8, no. 3, p. 154–162.

(26) DYMOND, T. S., HUGHES, F., AND JUPE, C. 1905 The influence of sulfates as manure upon the yield and feeding value of crops. *In* Jour. Agr. Sci., v. 1, p. 217–229.

(27) FEILITZEN, H. VON. 1913 Ueber die Verwendung der Schwefelblüte zur Bekämpfung des Kartoffelschorfes und als indirekes Dungemittel. *In* Fühling's Landw. Ztg., Bd. 62, p. 231–242.

(28) FRED, E. B., AND HART, E. B. 1915 The comparative effect of phosphates and sulfates on soil bacteria. Wis. Agr. Exp. Sta. Res. Bul. 35, p. 35–66.

(29) GALTSEV, P. E., AND YAKUSHKIN, I. V. 1913 Siernyi kolchedan i sul'faty zhelieza v ikh vliianii na fosforit. *In* 'Isv. Moskov. Selsk. Khoz. Inst. (Ann. Inst. Agron. Moscou), v. 19, no. 1, p. 223–232.

(30) GIANITTO, F. 1913 Contributo allo studio dello solfo come fertilizzante. *In* Bol. Quind. Soc. Agr. Ital., v. 17, p. 425–429. *Abs. in* Chem. Abs., v. 7, p. 1254.

(31) HART, E. B., AND PETERSON, W. H. 1911 Sulphur requirements of farm crops in relation to the soil and air supply. Wis. Agr. Exp. Sta. Res. Bul. 14, p. 21.

(32) HART, E. B., AND TOTTINGHAM, W. E. 1915 Relation of sulphur compounds to plant nutrition. *In* Jour. Agr. Res., v. 5, no. 6, p. 233–250.

(33) HEINZE, B. 1913 Die Steigerung des Bodenertrages durch den Schwefel. *In* Naturwissenschaften, Bd. 1, no. 5, p. 111–113.

(34) JANICAUD, W. 1914 Wirkt Schwefeldüngung wachstumsfördernd? *In* Gartenwelt, Bd. 18, Heft. 3, p. 29–32.

(35) KAPPEN, H., AND QUENSELL, E. 1915 Ueber die Umwandlungen von Schwefel und Schwefelverbindungen im Ackerboden, etc. *In* Landw. Vers. Stat., Bd. 86, p. 1–34.

(36) KOSSOVICH, P. S. 1913 Ueber den Kreislauf des Schwefels und chlors auf der Erde, etc. *In* Zhur. Opuitn. Agron. (Russ. Jour. Exp. Landw.), v. 14, p. 181–228.

(37) LIECHTI, P. 1913 Ueber die Wirkung des Schwefels auf das Pflanzenwachstum. *In* Mitt. Lebensan. Hyg., Bd. 4, Heft. 4, p. 267–268. *Abs. in* Chem. Abs., v. 8, no. 4, p. 774 (1914).

(38) LIERKE, E. 1913 Die Wirkung des in dem Boden gebrachten Schwefels. *In* Deut. Obstbau Ztg., Bd. 59, Heft 4, p. 75–76.

(39) LIESKE, R. 1912 Untersuchungen über die Physiologie denitrifizierender Schwefelbakterien. *In* Ber. Deut. Bot. Gesell., Bd. 30, General versamml., p. 12–22.

(40) LINT, H. C. 1914 The influence of sulfur on soil acidity. *In* Jour. Indus. Engin. Chem., v. 6, p. 747.

(41) LINT, H. C. 1914 Report of potato scab experiments. *In* N. J. Agr. Exp. Sta. 35th Ann. Rpt., p. 477–488.

(42) LINT, H. C. 1915 Report of potato scab experiments. *In* N. J. Agr. Exp. Sta., 36th Ann. Rpt., p. 375–381.

(43) LIPMAN, J. G. 1916 Sulfur on alkali soils. *In* Soil Sci., v. 2, no. 3, p. 205.

(44) LIPMAN, J. G., AND MCLEAN, H. C. 1917 Vegetation experiments on the availability of treated phosphates. *In* Soil Sci., v. 4, no. 4, p. 337–342.

(45) LIPMAN, J. G., AND MCLEAN, H. C. 1918 Experiments with sulfur-phosphate composts conducted under field conditions. *In* Soil Sci., v. 5, p. 243-250.

(46) LIPMAN, J. G., MCLEAN, H. C., AND LINT, H. C. 1916 The oxidation of sulfur in soils as a means of increasing the availability of mineral phosphates. *In* Soil Sci., v. 1, no. 6, p. 533–539.

(47) LIPMAN, J. G., MCLEAN, H. C., AND LINT, H. C. 1916 Sulfur oxidation in soils and its effect on the availability of mineral phosphates. *In* Soil Sci., v. 2, no. 6, p. 499–538.

(48) MCCONNELL, P. 1913 A Ruakura experiment. *In* Jour. Agr. (New Zeal.), v. 7, no. 3, p. 252–259.

(49) MCLEAN, H. C. 1916 Sulfur oxidation in soils as affecting the availability of mineral phosphates. Master's Thesis, Rutgers College.

(50) MCLEAN, H. C. 1917 Further studies on sulfur oxidation in soils as affecting the availability of mineral phosphates. Doctor's Thesis, Rutgers College.

(51) MAGNIEN, A. 1913 Experience sur l'emploi du soufre comme engrais, dans une culture des navets. *In* Jour. Soc. Nat. Hort. France, s. 4, t. 14, p. 54–56.

(52) MARÉS, H. 1858 Manuel pour le Soufrage des Vignes Malades. Paris.

(53) MARÉS, H. 1869 Des transformations que subit le soufre en poudre quand il est repandu sur le sol. *In* Compt. Rend. Acad. Sci. (Paris), t. 69, p. 974–979.

(54) MIÉGE, E. 1914 La question du soufre en agriculture. *In* Rev. Sci. (Paris), t. 52, s. 1. no. 25, p. 778–784.

(55) PETERSON, W. H. 1914 Forms of sulphur in plant materials and their variation with the soil supply. *In* Jour. Amer. Chem. Soc., v. 36, no. 6, p. 1290–1300.

(56) PETERSON, W. H. 1914 Sulphur in plant nutrition. *In* Wis. Agr. Exp. Sta. Bul. 240, p. 18–19.

(57) PFEIFFER, T., AND BLANCK, E. 1913 Beitrag zur Wirkung des Schwefels auf die Pflanzen-production, etc. *In* Landw. Vers. Stat., Bd. 83, p. 359–384.

(58) PFEIFFER, T., AND SIMMERMACHER, W. 1915 Beitrag zur Wirkung des Schwefels auf die Pflanzen-production. *In* Fühling's, Landw. Ztg., Bd. 64, Heft 9–10, p. 243–255.

(59) PITZ, W. 1916 Effect of elemental sulphur and of calcium sulphate on certain of the higher and lower forms of plant life. *In* Jour. Agr. Res., v. 5, no. 16, p. 771–780.

(60) RABATÉ, E. 1912 Emploi de l'acide sulphurique pour la destruction des mauvaises herbes dans les champs de ble. *In* Prog. Agr. Vit., t. 58, p. 568–572, 591–595, 629–636.

(61) REIMER, F. C. 1914 Sulphur fertilizer for alfalfa. *In* Pacific Rural Press, v. 87, no. 26, p. 717.

(62) ROBINSON, W. O. 1914 The inorganic composition of some important American soils. U. S. Dept. Agr. Bul. 122, p. 27.

(63) SABASHNIKOV, V. V. 1912 Novye opyty s' siernym' tsietom' kak udobreniem. *In* Zhur. Opuitn. Agron. (Russ. Jour. Exp. Landw.), v. 13, p. 817–822.

(64) SHEDD, O. M. 1913 The sulfur content of some typical Kentucky soils. Ky. Agr. Exp. Sta. Bul. 174, p. 269–306.

(65) SHEDD, O. M. 1914 The relation of sulfur to soil fertility. Ky. Agr. Exp. Sta. Bul. 188, p. 595–630.

(66) SHERBAKOFF, C. D. 1915 The after effect of sulfur treatment on soil. *In* Phytopathology, v. 5, no. 4, p. 219–222.

(67) THALAU, W. 1913 Die Einwirkung von im Boden befindlichen Sulfiten, von Thiosulphat und Schwefel auf das Wachstum der Pflanzen. *In* Landw. Vers. Stat., Bd. 82, Heft 3–4, p. 161–209.

(68) TOTTINGHAM, W. E. 1913 Effect of level of sulfur supply on plant growth. Wis. Agr. Exp. Sta. Bul. 228, p. 26.

(69) TRITSCHLER. 1913 Zur Frage der Schwefeldüngung. *In* Deut. Landw. Press, Bd. 40, Heft 33, p. 405.

(70) URBAN, J. 1913 Ueber die Wirkung der Schwefelblüte, auf das Wachstum der Zuckerrübe. *In* Ztschr. Zuckerindus. Bohmen, Bd. 37, Heft. 9, p. 441–444.

(71) VERCIER, J. 1912 Experience sur l'action fertilisante du soufre. *In* Prog. Agr. Vit., t. 58, p. 695–697.

(72) VERMOREL, V. 1914 Étude de l'action fertilisante du soufre sur la vigne. *In* Bul. Soc. Nat. Agr. France, t. 74, no. 1, p. 48–51.

(73) VERMOREL, V., AND DANTONY, E. 1913 Contribution à l'étude du soufre et des pyrites de fer employes comme engrais. *In* Engrais, t. 28, p. 1304–1306.

(74) VOELCKER, J. A. 1913 Sulfur as a fertilizer. *In* Jour. Roy. Agr. Soc. England, v. 74, p. 419.

(75) WHEELER, H. J., HARTWELL, B. L., AND MOORE, N. L. C. 1899 Upon the after effect of sulfur, when applied to soils for the purpose of preventing potato scab. *In* R. I. Agr. Exp. Sta. 12th Ann. Rpt., p. 163–167.

(76) ZOLLA, D. 1915 L'action fertilisante du soufre sur la vigne. *In* Rev. Gen. Sci., t. 26, p. 120–121.

TWENTY YEARS' WORK ON THE AVAILABILITY OF NITROGEN IN NITRATE OF SODA, AMMONIUM SULFATE, DRIED BLOOD AND FARM MANURES

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During the last twenty-five years the fertilizer industry in the United States has developed rapidly. From a comparatively small tonnage in the early nineties it has grown to more than 7 million tons in 1917.

As the industry has grown, the number of materials that go to make up the fertilizers has also increased greatly. Many by-products that were formerly allowed to go to waste are now carefully saved and worked up in the fertilizer factory. This is especially true of the nitrogenous materials, which, under normal conditions, form the most expensive part of the fertilizer.

The movement to save these waste materials containing nitrogen came none too early, for it was the depletion in the soil of this element that was largely responsible for the run-down and abandoned farms in the older sections of the United States. For this element, most crops show a quicker response than for any other, and conversely, a falling off in yield will come sooner with a deficiency of nitrogen than of any other element. A supply of available nitrogen aids the plant in getting a good start so that its leaves may begin early to elaborate food from the air and its roots may reach out for the water of the soil which holds in solution mineral plant-food.

Since nitrogen is supplied in many different forms, it at once becomes a question as to which of these is most efficient in crop production. Far too little attention has been given to this important question. Too often a certain material has been chosen because there was among farmers a general impression that this particular material was better than some other, when, as a matter of fact, there was no scientific basis for such conclusion. As an example, nitrogen from organic sources has been preferred by many because it was believed that organic matter thus supplied would be of great value in improving the physical condition of the soil, but in making this choice farmers overlooked the possibility of using a more readily-available material which would increase the crop residue sufficiently to more than make up for the small amount of organic matter contained in the few hundred pounds of dried blood, fish or tankage. Also, there is a widespread impression that the loss of nitrogen is greater when nitrates are used, than when organic nitrogen is used. But experiments both in this country and abroad show beyond a doubt that the

crop yields and the percentage of nitrogen recovered in the crop were greater (and hence the loss must have been less) when nitrates were used than when organic sources of nitrogen were used.

The question of availability of nitrogenous fertilizers began to receive serious consideration at several of the leading European experiment stations some 30 years ago and much valuable information has been accumulated by these stations.

About 10 years later the subject began to receive attention in this country and it is a satisfaction to find that the results obtained here are fairly in accord with the findings of the European investigators.

Fairly complete reviews of this early work have been given in recent publications (2, 3) and no attempt will be made here to cover this field.

The completion in 1917 of 20 years' work in which a comparison is made of the materials mentioned in the title of this paper would seem to justify the publication at this time of a brief summary of the work. A detailed account covering the first 15 years of this work has already been published (3). Much of this need not be repeated, but the results of the last 5 years are of value as confirming the earlier work.

EXPERIMENTAL

The work was originally outlined under the broad heading "Investigations Relative to the Use of Nitrogenous Materials," and this included: (a) a determination of the yield of dry matter and nitrogen in crops from soils receiving various treatments under controlled conditions; (b) the percentage of nitrogen in the crop as affected by the treatment; (c) the utilization of nitrogen in different materials; (d) the relative efficiency of nitrogen in different materials; (e) the residual effects of nitrogenous substances; (f) denitrification; and (g) the effect of special treatment on the income and outgo of nitrogen in the soil.

As the work has progressed, more and more attention has been given to the utilization and relative efficiency of nitrogen in different materials, and it is this phase of the work which is to receive consideration in this paper.

In order that the work might be under more perfect control, it was carried out in galvanized iron cylinders, open at both ends and having a diameter of 23½ inches and a depth of 4 feet. These cylinders were set in the ground so that about 2 inches remained above the ground level. Thus the contents of the cylinders are isolated so that the roots of the crops growing in them are prevented from getting mineral plant-food from outside sources. The subsoil is a gravelly sandy material such as occurs where the cylinders are located, but the top soil is a loam (Penn loam) brought from another source, an 8-inch layer of which was placed in each cylinder on top of the subsoil, each cylinder receiving the same weight of the thoroughly mixed soil.

When the work was begun all the soils were given a liberal treatment of lime in the form of ground limestone and with the exception of one series which

does not enter into this discussion, all have received annual dressings of acid phosphate and potassium chloride at the rate of 640 pounds and 320 pounds per acre, respectively. Thus nitrogen is made the limiting factor insofar as human control can provide. Various combinations of manure and fertilizer were arranged, but it is sufficient to report here only on the four nitrogenous materials mentioned in the title.

One series received the phosphoric acid and potash, but no nitrogen, in order that it might be used as a check. Thus if a certain amount of nitrogen is recovered in the crop from the nitrogen-treated cylinder, and it is desired to calculate the percentage of the applied nitrogen that was recovered, it is necessary first to deduct from the total amount of nitrogen recovered in the crop, the amount recovered from the check cylinder, and thus account for the soil nitrogen that the crop used.

It is at once obvious that this cannot be an absolutely correct method of determining the percentage recovered, since in those cylinders to which nitrogenous fertilizers have been applied, the plant will make a quicker start and the roots go farther in search of the nitrogenous materials of the soil than in the check cylinders where there is a pronounced deficiency of available nitrogen, and thus the check fails to be a *true* check. In this way it happens that the recovery may *apparently* be more than 100 per cent as shown in Series 8 B, for the years 1901 and 1910. However, there appears to be no way of overcoming this error so long as the work is carried out in the natural soil and if one starts with an artificial soil, other and more serious difficulties arise.

In this work no effort has been made to analyze the roots, since it would be well-nigh impossible to do this correctly, and even if it could be done the same error would be introduced. The roots and stubble are left just as under field conditions so that the residual effects of these may be observed.

To draw conclusions from 1 or 2 years of such work would be manifestly unfair, but when it is carried on for a period of 10 or 20 years, seasonal differences, differences due to the unequal decomposition of organic matter and differences due to slight errors, which are sure to creep in now and then, are largely smoothed out and results are obtained which can be accepted with a fair degree of confidence. The confidence in such results is strengthened when it is found that they check with similar work conducted in other places or even in other countries.

The work was started in these cylinders in the spring of 1898 with corn as the first crop in the rotation. Four 5-year rotations have been carried out as follows:

First Rotation		*Second Rotation*	
1898	Corn	1903	Corn
1899	Oats (millet)	1904	Oats (corn)
1900	Oats (corn)	1905	Oats (corn)
1901	Wheat	1906	Wheat
1902	Timothy (two cuttings)	1907	Timothy (two cuttings)

Third Rotation		*Fourth Rotation*	
1908	Corn	1913	Corn
1909	Oats (corn)	1914	Oats (corn)
1910	Oats (corn)	1915	Oats (corn)
1911	Rye and oats	1916	Wheat
1912	Timothy (two cuttings)	1917	Timothy (one cutting)

The corn following the oats is grown as a residual crop (without further addition of fertilizers) to utilize any nitrogen which the oat crop may have failed to get. All corn is planted thick and harvested as forage rather than as mature corn. Oats are harvested as oat-hay just before maturity, and wheat is harvested at maturity and saved as grain and straw.

Nitrogenous materials are applied for each main crop in the rotation as follows:

Cylinder 4B, farm manure, at the rate of 16 tons per acre.
Cylinder 8B, nitrate of soda, at the rate of 320 pounds per acre.
Cylinder 17B, ammonium sulfate, equivalent to 320 pounds of nitrate of soda per acre.
Cylinder 18B, dried blood, equivalent to 320 pounds of nitrate of soda per acre.

Thus a careful record is kept of the amount of nitrogen applied each year and of the yield of dry matter from each cylinder. From determinations of the amount of nitrogen in the dry matter the total amount of nitrogen removed by the crop each year is easily calculated.

YIELD OF DRY MATTER

The yield of dry matter under the four different treatments for the 20 years is shown in table 1, averages being given for two 10-year periods and also for the entire 20 years. For each 10-year period the yield has been largest with the manure, though it is less for the second 10-year period than for the first, which would indicate that with manure at the rate of 16 tons per acre the fertility of the soil is not being fully maintained. The lowest yield is from 18B where dried blood is used as the source of nitrogen. Here again the average yield is less for the second 10-year period than for the first. For plots 8B and 17iB, which receive nitrate of soda and ammonium sulfate, respectively, the average yields for the first 10 years are essentially the same for the two treatments, but for the second 10 years the average for the nitrate of soda treatment is considerably above that for the ammonium sulfate; furthermore, the average yield with ammonium sulfate is, like the yield with dried blood and farm manure, less for the second than for the first 10-year period. With the nitrate of soda, however, the figures are reversed, that is, the average yield for the second 10 years is somewhat above that for the first 10 years.

The question may well be raised as to why the average yields on 4B, 17B and 18B should be less for the second 10-year period than for the first, while the yield on 8B has been well maintained for the 20 years. Since phosphoric

acid and potash have been supplied each year in liberal amounts, and lime has been used at stated intervals, it would seem clear that the falling off in yield must be due to a deficiency of available nitrogen, notwithstanding the fact that cylinders 17B and 18B receive each year just as much nitrogen as 8B, while 4B receives more than two and one-half times as much as 8B.

Data presented heretofore, and which are confirmed by results hereafter to be presented, show that of the four materials, nitrate of soda is most effective in crop production, that is, the crop is able to utilize or win back a larger percentage of nitrogen in this form than in any of the other forms. With a given amount of nitrogen, therefore, the crop yield can be better maintained over a period of years by the use of nitrogen in the form of nitrate of soda than in the other forms, provided the soil is one that does not allow rapid leaching.

TABLE 1

Yield of dry matter with different nitrogenous materials

FIRST 10-YEAR PERIOD						SECOND 10-YEAR PERIOD					
Year	Check*	4B	8B	17B	18B	Year	Check*	4B	8B	17B	18B
	gm.	*gm.*	*gm.*	*gm.*	*gm.*		*gm.*	*gm.*	*gm.*	*gm.*	*gm.*
1898	291.1	467.1	393.9	401.0	341.8	1908	169.0	326.0	331.0	286.0	228.0
1899	146.6	354.1	184.5	190.5	186.3	1909	164.0	208.0	244.0	217.0	218.0
1900	238.1	387.2	317.0	310.1	307.9	1910	214.0	422.0	338.0	287.0	276.0
1901	126.0	342.2	331.0	300.0	239.4	1911	68.0	236.0	160.0	117.0	126.0
1902	86.2	147.8	150.9	143.9	115.6	1912	88.0	221.0	187.0	153.0	115.0
1903	160.3	315.0	183.0	291.0	216.0	1913	177.2	390.5	312.5	228.5	286.5
1904	118.7	262.0	170.0	167.0	160.0	1914	137.0	285.8	222.4	196.9	198.3
1905	125.7	262.0	226.0	209.0	191.0	1915	103.7	231.2	211.0	178.3	147.5
1906	98.3	316.0	244.0	226.0	144.0	1916	91.4	250.9	217.3	181.6	112.9
1907	107.3	237.0	168.0	133.0	172.0	1917	71.1	229.0	208.0	167.0	139.0
Average†	149.8	309.04	236.83	237.15	207.4	Average‡	128.3	280.04	243.12	201.23	184.72

* Phosphoric acid, potash and lime; no nitrogen.

† First 10 years.

‡ Second 10 years.

This apparently is what has happened in this case. With the gradual exhaustion of *soil* nitrogen, which was made available by the use of lime, and the failure of the ammonium sulfate, blood and manure to give back in the form of crops as large a proportion of the applied nitrogen as the nitrate of soda, the yields with the former became gradually less.

The fact that cylinder 4B gave the largest average yield through 20 years must not be taken as meaning that the treatment given this cylinder is necessarily the best or most effective. It will be remembered that this cylinder receives cow manure at the rate of 16 tons per acre annually, the cost of which would be much in excess of the cost of 320 pounds of nitrate of soda or its equivalent in ammonium sulfate or dried blood, and therefore the larger

yield does not necessarily mean an efficient use of the applied nitrogen. As a matter of fact, the work shows this to be the least efficient of the four forms.

PERCENTAGE OF NITROGEN RECOVERED IN THE CROPS

Reference has already been made to the method of calculating the percentage of nitrogen that is recovered in the crop. The recoveries for the four different treatments covering the 20 years are shown in table 2. The averages for the period are as follows:

4B	32.69 per cent (manure)
8B	62.42 per cent (nitrate of soda)
17B	47.48 per cent (ammonium sulfate)
18B	38.69 per cent (dried blood)

This means that of 100 pounds of nitrogen applied in the four different forms, approximately one-third, three-fifths, one-half, and two-fifths, respectively, are recovered or won back in the crop.[1] As has already been mentioned these figures agree quite closely with results reported from European countries, and they also confirm earlier work carried out at this Station.

But even so, they are not satisfying figures. We at once ask why there is this enormous loss of nitrogen and especially why the loss is so much greater with the organic materials than with the nitrate of soda and ammonium sulfate. If the loss is to be attributed to the leaching out of the materials, then it would seem that the figures should be reversed. Unquestionably, a certain amount of loss takes place in this way, but this cannot explain the loss of over two-thirds from the manure against a little more than one-third from nitrate.

It is well known that organic materials must undergo certain transformations in the soil before the nitrogen can become available, and it seems that during these transformations nitrogen as ammonia, nitrate or as elemental nitrogen must be lost in considerable quantities. As bearing on this it may be pointed out that Russell and Richards (5) have shown by laboratory experiments with manure that in addition to the loss of ammonia by volatilization there is still a loss amounting to 15 per cent or more of total nitrogen, and they have gone further and shown that during decomposition there is an evolution of gaseous nitrogen. This they believe completes the account of the loss. This loss, they claim, does not go on under wholly anaerobic or wholly aerobic conditions but under mixed anaerobic and aerobic conditions which arise when manure is being produced. They explain further that in the natural manure heap nitrogen is also lost as gaseous ammonia as well as in the form of nitrogen gas.

[1] Or if we assign to nitrate nitrogen a value of 100, then the relative availability of the four materials stands as follows:

Nitrate of soda	100.0
Ammonium sulfate	76.1
Dried blood	62.0
Manure	52.4

It is very probable that in a more limited way, similar changes take place when organic compounds are placed in the soil and that a part of the loss of nitrogen noted in our experiments must be thus accounted for. It is a well-known fact that when an organic substance like cottonseed meal or dried blood is mixed with soil and incubated in the laboratory for a few days, escaping ammonia may be detected, and from this it is a natural conclusion that when large quantities of organic matter are placed in the soil under natural conditions, some ammonia will be lost by volatilization, especially when the temperature and moisture conditions are favorable. This then, together with the evolution of gaseous nitrogen, would in part at least explain the heavy loss of nitrogen where manure was used at the rate of 16 tons per acre.

TABLE 2

Percentage of nitrogen recovered from different materials

FIRST 10-YEAR PERIOD					SECOND 10-YEAR PERIOD				
Year	4B	8B	17B	18B	Year	4B	8B	17B	18B
1898	28.15	63.75	66.06	58.18	1908	16.97	42.77	24.20	27.38
1899	51.48	48.45	58.27	44.58	1909	18.25	80.64	54.94	49.04
1900	36.18	77.55	69.47	57.25	1910	54.74	110.74	62.12	51.22
1901	41.78	110.26	91.91	68.71	1911	20.98	64.10	48.46	41.59
1902	11.48	32.06	23.64	14.32	1912	29.11	49.16	27.45	10.96
1903	20.20	30.84	34.38	20.97	1913	27.63	32.92	15.50	40.26
1904	38.91	46.19	39.26	33.68	1914	52.46	74.35	67.86	56.55
1905	30.10	68.77	56.05	34.01	1915	32.13	64.10	52.53	48.12
1906	44.94	81.81	30.80	24.78	1916	36.60	68.96	57.53	20.26
1907	33.85	45.10	27.47	42.48	1917	27.95	55.77	41.75	29.41
Average *	33.71	60.48	49.73	39.90	Average†	31.68	64.35	45.23	37.48
Average ‡	32.69	62.42	47.48	38.69					

* First 10 years.

† Second 10 years.

‡ Twenty years.

A discussion of this subject would not be complete without a brief reference to the effect of cultivation on nitrogen losses.

Shutt[2] for example has shown that when the prairie soils of Saskatchewan were left undisturbed the loss of nitrogen was slight, but as soon as cultivation was commenced losses set in.

Russell (4) refers further to losses of nitrogen as follows:

> One of the Broadbalk wheat plots receives annually 14 tons of farm-yard manure per acre containing 200 pounds of nitrogen. Only a little drainage can be detected and there is no reason to suppose that any considerable leaching out of nitrates occurs, but the loss of nitrogen is enormous amounting to nearly 70 per cent of the added quantity.
>
> The condition for this decomposition appears to be copious aeration, such as is produced by cultivation and the presence of large quantities of easily decomposable organic matter.

[2] Cited by Russell (4).

Now these are precisely the conditions of intensive farming in old countries and of pioneer farming in new lands, and the result is that the reserves of soil and manurial nitrogen are everywhere being depleted at an appalling rate.

Russell refers to the recuperative actions that are going on, but says; "One of the most pressing problems at the present time is to learn how to suppress this gaseous decomposition and to direct the processes wholly into the nitrate channels."

In a paper on the nitrate content of cultivated and uncultivated soils, Blair and McLean (1), have called attention to the loss of nitrogen from cultivated soils and also to the low recovery from nitrogen applied as organic materials. They point out that land under cultivation is gradually being depleted of its store of nitrogen even when nitrogenous fertilizers are applied each year and that the average recovery of nitrogen applied in the form of fish scrap for a period of nine years, was only 36.36 per cent.

With the same nitrogen treatment soils allowed to run wild just about maintained their nitrogen content, while the carbon content of these soils was slightly increased.

The recovery of nitrogen in the four different treatments for the 20 years is shown by the curves in figure 1. A study of these curves shows that the high points are generally reached in either the first or second year of oats, and in the wheat year, while the low points occur almost invariably in the corn and timothy years. It is not entirely clear whether this is a seasonal variation or a crop characteristic.

It is certain, however, that the utilization of the residual nitrogen by the corn crops which follow the oats, helps to explain the high recovery for the years when oats are grown.

CONCLUSIONS

In a 5-year rotation on Penn loam soil well supplied with phosphoric acid, potash and lime, crop yields were better maintained over a period of 20 years with nitrate of soda at the rate of 320 pounds per acre than with an equivalent amount of ammonium sulfate or dried blood. For several years the latter gave results about on a par with the nitrate, but an average of the second 10-year period shows a considerable falling off with these materials as compared with the nitrate. This is no doubt due in part to the fact that the nitrate, being immediately available, gives the plant an early start which tends to keep it in the lead, and to the further fact that in the transformation of the ammonium salt and the organic material into nitrates, there is a considerable loss of nitrogen, possibly as ammonia gas or gaseous nitrogen or both. The loss cannot all be attributed to a leaching out of the materials, even though the nitrification of ammonia and organic residues may go on throughout a large portion of the year.

In the above-mentioned rotation cow manure at the rate of 16 tons per acre gave somewhat larger yields than nitrate of soda, but the increased yields were not sufficient to justify the increase in the cost of nitrogen.

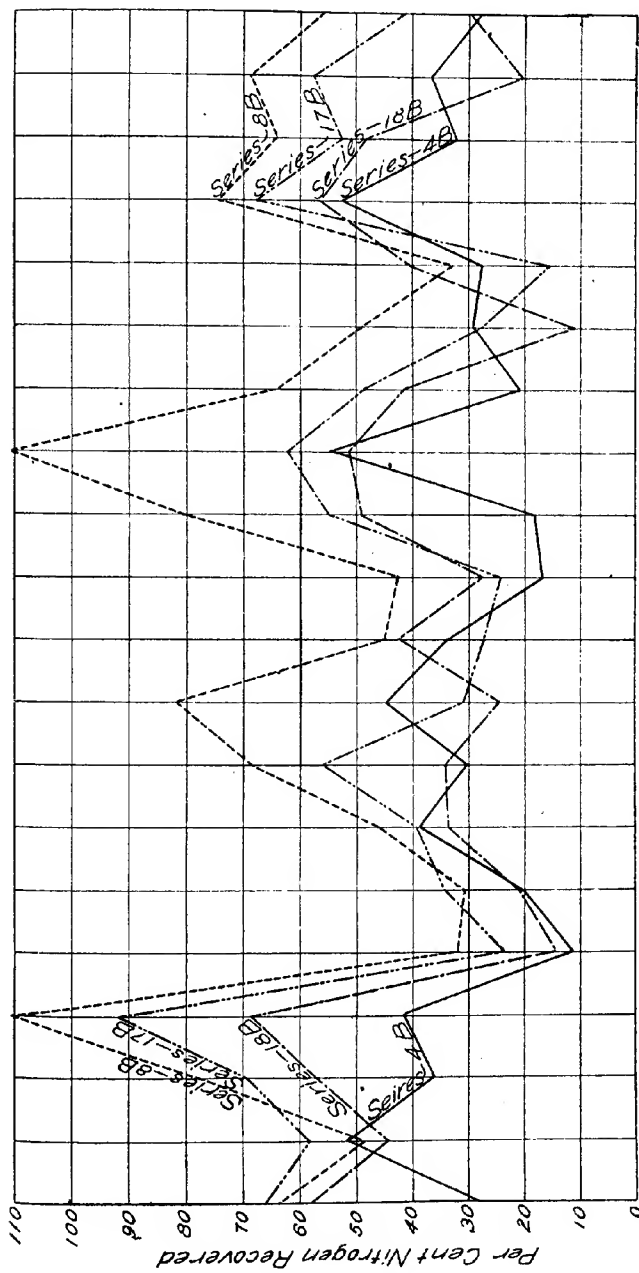

FIG. 1. DIAGRAM SHOWING RECOVERY OF NITROGEN FOR THE FOUR DIFFERENT TREATMENTS OVER A PERIOD OF 20 YEARS

Furthermore, the average yield with the manure was less for the second 10-year period than for the first, while the reverse is true with the nitrate of soda. Thus it is shown that with 16 tons of manure per acre annually, the crop yield is not being maintained, while with nitrate of soda at the rate of 320 pounds per acre annually it is increasing slightly, as shown by the average for the second 10-year period.

The percentage of nitrogen recovered in the crop was greater with the nitrate than with any of the other materials, the 20-year average being as follows:

	per cent
Nitrate of soda	62.42
Ammonium sulfate	47.48
Dried blood	38.69
Cow manure	32.69

The average recovery with nitrate for the second 10-year period was 64.35 per cent as against 60.48 per cent for the first 10-year period, whereas the average recovery with the ammonium sulfate, dried blood and manure were all less for the second 10-year period than for the first.

This is in agreement with the crop yields, and indicates a diminishing efficiency for the ammonium sulfate, blood and manure, and a gradual increase in efficiency for the nitrate of soda.

The work shows that when properly used nitrate of soda alone as a source of nitrogen may be depended upon to maintain crop yields over a long period, and that a given amount of nitrogen in this form is more effective than an equivalent amount in the form of ammonium sulfate, or organic materials.

Its effect is to produce larger crops per unit of nitrogen, and these crops, in turn, leave behind in the soil larger crop residues, and with carbonate of lime to aid in their decomposition these furnish a sufficient supply of organic matter to keep the soil in good physical condition.

REFERENCES

(1) Blair, A. W., and McLean, H. C. 1917 Total nitrogen and carbon in cultivated land and land abandoned to grass and weeds. *In* Soil Sci., v. 4, no. 4, p. 283–294.

(2) Coleman, D. A. 1917 The influence of sodium nitrate upon transformations in soils with special reference to its availability and that of other nitrogenous manures. *In* Soil Sci., v. 4, no. 5, p. 345–432.

(3) Lipman, J. G., and Blair, A. W. 1916 Investigations relative to the use of nitrogenous plant foods, 1898–1912. N. J. Agr. Exp. Sta. Bul. 288.

(4) Russell, E. J. 1915 Soil Conditions and Plant Growth, new ed., p. 83, Longmans, Green and Co., New York.

(5) Russell, E. J., and Richards, E. H. 1917. The changes taking place during the storage of farmyard manure. *In* Jour. Agr. Sci., v. 8, p. 495–563.

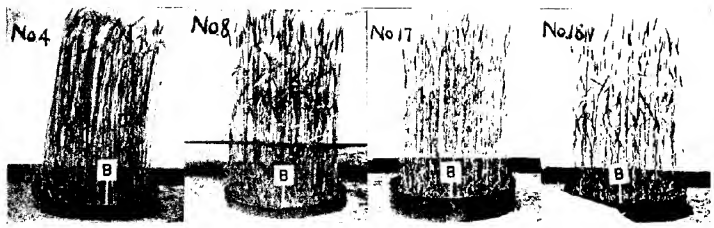

FIG. 1. WHEAT 1916

No. 4, cow manure, 16 tons per acre; No. 8, nitrate of soda, 320 pounds per acre; No. 17, ammonium sulfate equivalent to the nitrate; dried blood equivalent to the nitrate.

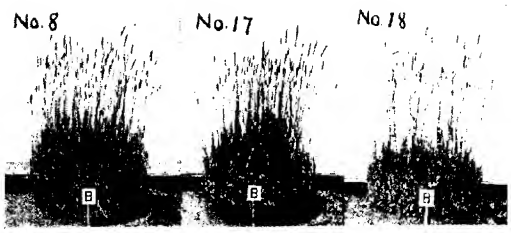

FIG. 2. TIMOTHY 1917

No. 8, nitrate of soda as above; No. 17, ammonium sulfate as above; No. 18, dried blood as above. (No photograph secured of No. 4 in 1917.)

A METHOD FOR THE COUNTING OF CERTAIN PROTOZOA IN THE SOIL

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With the recent developments in soil protozoology, various methods for counting protozoa have been devised as well as means of cultivation. The chief methods in use at the present time are five in number, namely: (a) the direct counting of a drop; (b) the dilution method of Rahn (6); (c) the agar-plating method of Killer (2); (d) the use of the standard loop of Müller (5); (e) the use of the blood counting apparatus as described by Kopeloff, Lint and Coleman (4). This last is perhaps the most recent. A review of the above methods is given by Kopeloff and Coleman (3).

In connection with our work on soil protozoa the last-named method seemed very satisfactory, although difficulties were encountered under certain conditions. The authors found the accuracy of the method to be impaired (a) when the number of organisms in the sample fell below four hundred per cubic centimeter of suspension, and (b) when by any chance soil particles, the diameter of which was greater than 0.1 mm., had been carried into the chamber. The probability of error due to settling while taking the sample will be discussed and demonstrated in the data.

To meet our conditions the following method was devised and was based on the principles of the Sedgwick-Rafter cell,[1] commonly used in the estimation of plankton in water examination, in conjunction with vital stain and a semi-solid diluent.

APPARATUS

The cell was of the type described by Whipple (7). The cell used was made by E. A. Thompson, of Amherst, and consisted of a large slide 75 by 25 mm. upon which was cemented a brass plate exactly 1 mm. in thickness, having in the center a rectangular opening 50 by 20 mm. A cover slip of the same size was used, thus giving a cell of 1 cc. capacity.

MICROSCOPES

In the course of the work two microscopes were used, a Leitz and a Bausch and Lomb binocular. The Leitz was used with the no. 3 objective and no. 4 ocular, this combination having the following physical constants:

[1] The use of this cell was suggested by Mr. Arthur W. Walker of the Northwestern University Medical School.

Numerical aperature	0.30
Initial magnification	10.3 mm.
Working distance	5.5 mm.
Diameter of visible field	2.1 mm.
Equivalent focus	16.2 mm.
Magnification	103×

The Bausch and Lomb binocular was used in the greater part of the work. In all cases the oculars were 10× and the objectives either 24-mm. or 40-mm. The former gave a magnification of 74× and the latter a magnification of 34×. With the first-named the diameter of the visible field was 2 mm., with the latter it was 4.25 mm.

CHEMICALS

The vital stain was the indicator phenolsulphonephthalein. It was procured in ampoules as sold for injection for the renal efficiency test and used without altering its condition.

The diluent consisted of a 4 per cent solution of gelatine (Gold Label), the reaction of which had been adjusted to P_H 8 approximately by the addition of N/5 NaOH.

METHOD OF PROCEDURE

For test work the organisms were cultivated in mannite solution (1). Samples of soil were put in the medium, usually about 5 gm. of soil to 100 cc. of medium and then the mixture was gently shaken. This was allowed to stand for two to three weeks at room temperature and usually at the end of this time was found to contain an abundance of organisms.

The counting, unless otherwise designated, was carried on in the following manner:

The cell was thoroughly cleaned with water and wiped with xylene. By means of a platinum loop the bottom was covered with a thin layer of the phenolsulphonephthalein solution. This was allowed to dry and the culture, previously diluted with the alkaline gelatine solution, was run in after the manner described by Whipple (7). The mixture was then counted and from these data in the usual manner the total number of organisms per cubic centimeter of suspension was determined,

In the work to determine the relation of time of settling to the total number of organisms, the same procedure was followed throughout with the exception that 1 gm. of sand was added before the dilution was made up to 10 cc. and the whole gently shaken until the sand was evenly distributed. The time of taking the samples is given in the data.

DATA

I. Count made by Sedgwick-Rafter method without vital stain or sand

[a]A culture of large ciliates was taken and the counts were made with the Bausch and Lomb binocular with 10× oculars and 40-mm. objectives. The number of fields to total area of cell was 81.9.

COUNT	FIELDS										AVERAGE	TOTAL	D*	D^2†
	1	2	3	4	5	6	7	8	9	10				
1	1	4	5	2	2	1	1	5	3	4	2.8	229.0	36.0	1,296.0
2	2	3	0	2	2	2	2	5	3	2	2.3	188.0	5.0	25.0
3	1	2	0	1	4	1	1	7	1	4	2.2	180.0	13.0	169.0
4	1	6	1	4	4	3	0	2	0	1	2.2	180.0	13.0	169.0
5	1	3	2	4	2	1	3	4	3	2	2.3	188.0	5.0	25.0
Sum												965.0		1684.0
Mean												193.0		

* The deviation from the mean.

† Square of the deviation from the mean.

$$E_s = \pm 0.6745 \sqrt{\frac{1684.0}{5}}$$

$$= \pm 12.37$$

Percentage of error = 12.37 ÷ 193.0 × 100 = 6.41

The probable error for a single counting is calculated according to the usual formula:

$$E_g = \pm 0.6745 \sqrt{\frac{\Sigma D^2}{n}}$$

where E_s is the error of single variate,
0.6745 is a constant for standard deviation.
ΣD is the sum of the squares of the deviations from the mean,
and "n" is the number of variates.

It will be noted that in the above results there is considerable error. This seems to be due to the fact that the protozoa were not defined and therefore some may not have been seen.

II. A count of small flagellates as in I

Unstained and without sand. A dilution of 1:20 was used. The microscope was binocular with 10× oculars and 24-mm. objectives. There were 318.5 fields, in the total area of the cell.

COUNT	FIELDS										AVERAGE	TOTAL/20	D	D^2
	1	2	3	4	5	6	7	8	9	10				
1	2	2	2	2	3	4	1	0	0	1	1.7	541.4	19.14	366.33
2	1	2	3	0	3	3	1	1	0	0	1.4	445.9	114.64	13,142.30
3	1	4	2	0	1	0	1	3	3	2	1.7	541.4	19.14	366.33
4	4	3	2	0	3	1	2	1	2	0	1.8	573.3	12.76	162.33
5	4	1	2	3	2	2	2	3	1	2	2.2	700.7	140.16	19,648.00
Sum												2,803.7		33,685.7
Mean												560.54		

$$E_s = \pm 0.6745 \sqrt{\frac{33685.78}{5}}$$

$$= \pm 55.35$$

Percentage of error = 55.35 ÷ 560.54 × 100 = 9.87

The results indicate that the probable error increases considerably in this case. It may be due to the size of flagellates, and also to higher magnification which means a small visible field.

III. A count of large ciliates with Sedgwick-Rafter method and the use of vital stain

Binocular used with 10× oculars and 40-mm. objectives; dilution 1: 1.

COUNT	FIELDS										AVERAGE	TOTAL	D	D^2
	1	2	3	4	5	6	7	8	9	10				
1	3	2	1	1	1	2	1	4	0	0	1.5	246	4	16
2	2	1	2	1	2	1	1	3	0	2	1.5	246	4	16
3	2	2	2	1	0	2	1	0	2	2	1.4	230	12	144
4	1	2	1	0	3	0	1	2	3	2	1.5	246	4	16
Sum												968		192
Mean												242		

$$E_s = \pm 0.6745 \sqrt{\frac{192}{4}}$$

$$= \pm 4.6710$$

Percentage of error = 4.6710 ÷ 242 × 100 = 1.93

Calculating from these data it may be seen that the totals check very closely and the percentage of error is reduced considerably. In the experimental work it was found that the organisms were clearly defined.

IV. Count of small flagellates using vital stain but no sand

The binocular was used with 10× oculars and 24-mm. objectives. Suspension was diluted 1: 20.

COUNT	FIELDS										AVERAGE	TOTAL/20	D	D^2
	1	2	3	4	5	6	7	8	9	10				
1	4	3	2	0	2	2	1	0	2	2	1.8	573.3	19.14	366.3
2	2	2	2	3	0	1	1	0	3	3	1.7	541.4	2.76	7.6
3	1	3	0	3	0	3	2	1	3	3	1.9	605.1	50.94	2,594.9
4	5	2	1	1	1	2	0	3	1	1	1.7	573.3	19.14	366.3
5	3	1	1	2	3	2	1	1	0	1	1.5	477.7	76.46	5,846.0
Sum												2,770.8		9,181.1
Mean												554.16		

$$E_s = \pm 0.6745 \sqrt{\frac{9181.1}{5}}$$

$$= \pm 28.79$$

Percentage of error = 28.79 ÷ 554.16 × 100 = 5.21

Calculating as in the case of III we find a lower percentage of error than in the unstained preparations, although it is higher than in III. There seems to be but little doubt that this is due to the use of the stain.

V. Indicating the influence of sand upon the counting during the time of settling

A culture of large ciliates was used in connection with the proposed method and samples of 1 cc. were taken at 5-minute intervals starting with the sand, 60–80 mesh, still in suspension. Binocular with 10× oculars and 40-mm. objectives were used.

COUNT	FIELDS										AVERAGE	TOTAL	D	D^2
	1	2	3	4	5	6	7	8	9	10				
1	2	1	1	1	2	3	2	1	3	1	1.7	139.0	83.4	6,955.60
2	0	1	2	1	0	1	0	1	0	3	0.9	74.0	18.4	337.79
3	0	0	0	2	1	0	0	0	1	0	0.4	33.0	22.6	510.76
4	0	0	0	1	0	0	1	0	0	0	0.2	16.0	39.6	1,568.20
5	1	0	0	1	0	0	0	0	0	0	0.2	16.0	39.6	1,568.20
Sum												278.0		10,940.55
Mean												55.6		

$$E_s = \pm 0.6745 \sqrt{\frac{10940.55}{5}}$$

$$= \pm 31.55$$

Percentage of error = 31.55 ÷ 55.6 × 100 = 56.87

Calculations show the effect of time of settling in relation to the total number of organisms per cubic centimeter and the error resulting from this cause is very large, as indicated above.

VI. Check count on V

Same organisms and technique employed as in the case of V. In place of one sample of 10 cc., in this case 5 tubes were made up as in V and all were shaken simultaneously and the samples taken as noted above. Thus an error from change of dilution was avoided.

COUNT	FIELDS										AVERAGE	TOTAL	D	D^2
	1	2	3	4	5	6	7	8	9	10				
1	2	1	1	2	1	1	3	2	1	1	1.5	123	74	5,476
2	2	1	0	0	0	1	1	1	1	1	0.8	65	16	256
3	0	1	1	0	0	0	1	0	1	0	0.4	33	16	256
4	0	0	0	1	0	0	0	0	0	1	0.2	16	33	1,089
5	0	0	0	0	0	0	0	0	0	1	0.1	8	41	1,681
Sum												245		8,758
Mean												49		

$$E_s = \pm 0.6745 \sqrt{\frac{8758}{5}}$$

$$= \pm 28.23$$

Percentage of error = 28.23 ÷ 49 × 100 = 57.62

Here the result of the preceding experiment is duplicated, indicating the very high probable error.

VII. Time of settling in relation to number of small flagellates

The tubes were used in the manner described under VI. One gram of sand was placed in each tube and the total volume made up to 10 cc. with a mixture consisting of a 1:5 dilution of the culture. The objectives used on the binocular were 24-mm.

COUNT	FIELDS										AVERAGE	TOTAL/5	D	D^2
	1	2	3	4	5	6	7	8	9	10				
1	2	9	12	4	8	9	4	4	3	7	6.2	1,974.6	923.6	853,005
2	8	3	5	2	3	5	3	3	3	2	3.7	1,178.4	127.4	16,268
3	4	3	5	3	2	2	1	3	3	2	2.8	891.8	159.2	26,539
4	4	3	3	5	1	1	1	1	1	2	2.2	700.6	350.4	125,860
5	2	2	1	1	1	3	2	1	2	1	1.6	509.6	541.4	293,110
Sum												5,255.0		1,314,782
Mean												1,051.0		

$$E_s = \pm 0.6745 \sqrt{\frac{1314782}{5}}$$

$$= \pm 345.88$$

Percentage of error = 345.88 ÷ 1051.0 × 100 = 32.09

The above shows quite clearly that small flagellates are affected in much the same way as the ciliates.

VIII. Check count on VII

Same technique as VII, except that a 1:10 dilution was used.

COUNT	FIELDS										AVERAGE	TOTAL/5	D	D^2
	1	2	3	4	5	6	7	8	9	10				
1	1	3	4	4	4	2	2	2	0	1	2.3	1,465	726.2	527,375
2	2	1	2	1	2	1	3	0	1	0	1.3	828	89.2	7,966
3	1	1	1	2	1	1	0	1	1	1	1.0	637	101.8	10,366
4	1	0	1	0	1	1	0	1	1	1	0.7	446	292.8	85,732
5	1	1	0	0	1	0	2	0	0	0	0.5	318	420.8	177,050
Sum												3,694		808,489
Mean												738.8		

$$E_s = \pm 0.6745 \sqrt{\frac{808489}{5}}$$

$$= \pm 27.23$$

Percentage of error = 27.23 ÷ 738.8 × 100 = 36.69

This count seems to check the results found in VII.

SUMMARY AND CONCLUSIONS

Percentage of probable error

LARGE CILIATES				SMALL FLAGELLATES			
Without sand		With sand		Without sand		With sand	
−Stain	+ Stain	- Stain	+ Stain	- Stain	+ Stain	- Stain	+ Stain
6.5	1.93*	56.87	57.62	9.87	5.21*	32.09	36.69

* Percentage of probable error for the proposed method obtained under the conditions described previously.
− signifies without stain.
+ signifies with stain.

It is the authors' experience that the above data seem to indicate several interesting points, namely:

1. The use of a vital stain aids materially in defining the organism.
2. The use of the semi-solid diluent inhibits the rapid motion,
3. The results of a count are seriously affected when the soil is allowed to settle.
4. The soil particles do not seem to affect the accuracy of the counts, especially when the vital stain is used.
5. The probable error seems to vary inversely with the size of the organisms and directly with the magnification.

The method is presented, not with the intention of supplanting methods already in use, but rather with the idea of supplementing certain methods which, within definite limits, give excellent results.

The method has its limitations as the authors well realize. The cell does not permit a magnification that will include all the protozoa in the soil, neither does it allow for the possibility of the organisms being concealed by the soil particles. Nevertheless, the data given above seem to show that under the stated conditions gratifying results were obtained.

The advantages of the method may be summed up in the following statements:

1. The sample counted consists of a comparatively large amount of soil dilution, therefore allowing the counting to be done while the soil is in suspension.
2. The vital stain brings the organisms in the field into prominence.
3. The use of the semi-solid diluent prevents rapid motion and lessens the chance of error.
4. The procedure is simple and quick.
5. The apparatus is inexpensive.

REFERENCES

(1) GILTNER, W., ET AL. 1916 Laboratory Manual in General Microbiology, p. 354. John Wiley and Sons, New York.

(2) KILLER, J. 1913 Die Zahlung der Protozoen im Boden. *In* Centbl. Bakt. (etc.), Abt. 2, Bd. 37, No. 17–21, p. 521–534.

(3) KOPELOFF, N. AND COLEMAN, D. A. 1917 A review of investigations in soil protozoa and soil sterilization. *In* Soil Sci., v. 3, no. 3, p. 197–269.

(4) KOPELOFF, N., LINT, H. C., AND COLEMAN, D. A. 1915 New methods in soil protozoology. *In* Science, n. s., v. 42, no. 1078, p. 284–286.

(5) MÜLLER, P. T. 1912 Über eine neue, rasch arbeitende Methode der bakteriologischen Wasseruntersuchung und ihre Anwendung auf die Prüfung von Brünnen und Filterwerken. *In* Arch. Hyg., Bd. 75, Heft 4–5, p. 189–223.

(6) RAHN, O. 1913 Methode zur Schätzung der Anzahl von Protozoen im Boden. *In* Centbl. Bakt. (etc.), Abt. 2, Bd. 36, No. 15–18, p. 419–421.

(7) WHIPPLE, G. C. 1914 Microscopy of Drinking Water, Ed. 3, p. 34.

SULFOFICATION IN RELATION TO NITROGEN TRANSFORMATIONS

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At present considerable interest is taken in the use of sulfur in agriculture. Experimental evidence has been obtained which indicates that finely-ground sulfur added to some soils has favorably influenced plant growth; and that the acidity produced by the process of sulfur oxidation has a remarkable solvent action on mineral plant nutrients, including phosphates, thereby converting them into more available forms.

In considering the possibilities of elementary sulfur as a means for either directly or indirectly increasing crop production, some questions arise as to the effect of the acidity produced on nitrogen transformations in acid and basic soils. Preliminary data pertaining to the relations between sulfofication and the production of ammonia and nitrates under controlled conditions are presented.

EXPERIMENTAL PROCEDURE

Description of soils

The soils used in the experimental work were representative of three distinct types and provided for wide variations in content of organic matter and basicity.

The silt loam soil which is deficient in calcium carbonate and organic residues has a total lime requirement of approximately 4000 parts of calcium carbonate per million. The black clay used is decidedly basic in reaction, although it contains only 300 parts of calcium carbonate per million.

The peat was thoroughly decomposed and required approximately 10,000 parts of calcium carbonate per million to satisfy its requirements for base.

Five hundred-gram portions of the soils reduced sufficiently to pass through a 2-mm. sieve were used as a basis for the different treatments.

Soil treatment

The additions where made were flowers of sulfur, casein, rock phosphate, and calcium carbonate. Sulfur and rock phosphate were supplied at the rate of 1000 parts per million; and the quantity of casein added furnished 500 parts of nitrogen per million.

For the purpose of studying the effect of a supply or deficiency of a readily salifiable base on the processes involved, the additions of calcium carbonate provided for different degrees of basicity. In the case of the acid silt loam the largest addition of calcium carbonate, 4000 parts per million, was slightly in excess of the amount necessary to satisfy completely the soil's requirement; one-half and one-fourth this amount also were added to certain mixtures of the silt loam soil.

The natural basicity of the black clay was increased by 1000 and 500 parts of calcium carbonate per million. The largest addition of calcium carbonate to the decidedly acid peat was 10,000 parts per million and the smallest 2000.

When the materials were mixed with the soils, water was added to satisfy 60 per cent of their water-holding capacity. The several mixtures were transferred to quart jars and placed in a darkened cabinet, the temperature of which was maintained at 28° to 30°C.

The total weights of the jars and contents were obtained at the beginning of the period and weighings made at regular intervals when the water lost by evaporation was replaced; weighings were made and water added every fourth day.

Optimum conditions for aeration prevailed, as the containers were uncovered and the mixtures stirred two days after each addition of water required to adjust their water content.

Fifty different mixtures were included in the experiment, duplicate jars of each being prepared on the same date. The interval between the beginning of the experiment and the time when the transformations which had occurred were determined was 17 weeks.

ANALYTICAL METHODS

At the end of the experimental period the amount of sulfur as sulfate, of nitric and ammoniacal nitrogen, the acidity and the alkalinity were determined in the water extract of the mixtures, according to the following methods.

Solution

At the end of the incubation period the contents of the jars were emptied into a large porcelain evaporating dish, thoroughly mixed, and returned to the jars as quickly as possible to prevent loss of weight by evaporation. A weighed portion of the mixture corresponding to 400 gm. of dry soil was transferred to wide-mouth bottles having a capacity of 4000 cc., and water added so that the total volume in contact with the soil was 2500 cc.

The extraction was continued through a period of 14 hours with shaking at regular intervals during the first two hours. Filtration was made through a battery of Berkfeld filters employing pressure secured from an automatically controlled pump. The first portion of the filtrate was discarded. Approximately 2000 cc. of solution were secured in each case.

Nitrogen as ammonia in water solution

A 200-cc. portion of the filtered solution was transferred into a Kjeldahl flask and a half gram of freshly calcined magnesium oxide added. The ammonia distilled off was passed through a second and smaller Kjeldahl flask used as a scrubber and from this into a 500-cc. Erlenmeyer flask made of Pyrex glass. All connecting tubing was made of Pyrex glass. The liberated ammonia was absorbed in 1/10 normal acid and titrated with 1/20 normal alkali, methyl red being used as an indicator.

Nitrate nitrogen

Nitrogen in this form was determined by the Devarda reduction method as modified by Allen (1).

The solution remaining after distilling off ammonia with magnesium oxide was diluted with 200 cc. of distilled water, free from ammonia and nitrates, 2 cc. of 50 per cent sodium hydroxide solution were added and the contents of the flask boiled for 20 minutes. The flasks were then cooled, 2 gm. of Devarda's alloy added, and at once connected to the same apparatus as was used for the ammonia determination, and boiled for 40 minutes.

Sulfates

Water-soluble sulfates were determined gravimetrically in 200-cc. aliquots of the filtered solution after first acidifying with hydrochloric acid and boiling to decompose carbonates before the addition of barium chloride.

Acidity and alkalinity

The acidity or alkalinity of the water extract of the mixtures expressed in terms of sulfuric acid or calcium carbonate was determined by titrating a 100-cc. aliquot of the solution as follows:

Ten cubic centimeters of N/50 H_2SO_4 were added, the solution boiled and cooled, and then titrated with N/50 NaOH solution, phenolphthalein being used as an indicator.

SULFUR OXIDATION

The results for soluble sulfur as sulfates extracted by water from the differently treated soils are included with the data for nitric and ammoniacal nitrogen, and acidity and basicity of the water solutions, in the accompanying tabulations.

The amounts of water-soluble sulfates obtained from those mixtures in which no sulfur was included were practically the same. This indicates that a greater proportion of the natural sulfur supply has been oxidized in the acid silt loam than in the basic clay, since the total sulfur content of the basic clay

TABLE 1

Sulfur and nitric and ammoniacal nitrogen in acid silt loam. Data expressed as parts per million of soil

TREATMENT	$CaCO_3$ ADDED	SULFUR	NITRIC NITROGEN	AMMONIACAL NITROGEN	ACIDITY AS H_2SO_4	ALKALINITY AS $CaCO_3$
None		20	43	0	0	9
Floats		18	77	0	0	25
Floats	4,000	52	116	0	0	212
None	4,000	58	103	0	0	140
Casein		44	200	132	61	0
Casein	4,000	56	440	0	0	56
Casein, floats		32	186	55	24	0
Casein, floats	4,000	59	400	11	0	87
Sulfur	4,000	671	88	0	0	12
Sulfur	2,000	675	22	33	52	0
Sulfur	1,000	710	11	46	223	0
Sulfur		718	8	33	511	[illegible]
Sulfur, floats	4,000	524	96	10	0	75
Sulfur, floats	2,000	553	29	38	30	0
Sulfur, floats	1,000	589	18	43	196	0
Sulfur, floats		775	12	48	666	0
Sulfur, casein	4,000	661	295	4	18	0
Sulfur, casein	2,000	808	92	241	76	0
Sulfur, casein	1,000	740	33	208	200	0
Sulfur, casein		725	10	328	144	0
Sulfur, floats, casein	4,000	646	296	88	Neutral	
Sulfur, floats, casein	2,000	752	98	254	79	0
Sulfur, floats, casein	1,000	683	44	208	128	0
Sulfur, floats, casein		716	44	288	159	0

TABLE 2

Sulfur and nitric and ammoniacal nitrogen in basic black clay. Data expressed as parts per million of soil

TREATMENT	$CaCO_3$ ADDED	SULFUR	NITRIC NITROGEN	AMMONIACAL NITROGEN	ALKALINITY AS $CaCO_3$
None		38	142	11	90
Floats		31	141	11	93
None	1,000	38	143	11	106
Floats	1,000	35	143	0	175
Sulfur		656	107	11	78
Sulfur, floats		790	144	10	78
Sulfur	1,000	811	145	11	43
Sulfur	500	707	146	10	93
Sulfur, floats	1,000	760	158	11	46
Sulfur, floats	500	723	145	16	90

TABLE 3

Sulfur and nitric and ammoniacal nitrogen in acid peat. Data expressed as parts per million of soil

TREATMENT	$CaCO_3$ ADDED	SULFUR	NITRIC NITROGEN	AMMONIACAL NITROGEN	ACIDITY AS H_2SO_4
None		118	255	259	353
Floats		108	210	252	412
None	10,000	108	556	122	137
Floats	10,000	87	560	115	39
Sulfur		937	105	357	294
Sulfur, floats		952	100	392	353
Sulfur	10,000	818	112	399	274
Sulfur	5,000	942	80	406	294
Sulfur	2,000	981	58	398	372
Sulfur, floats	10,000	1025	108	318	235
Sulfur, floats	5,000	935	98	420	118
Sulfur, floats	2,000	881	105	403	235

is about 500 parts per million and the silt loam contains 230. Apparently, conditions are more favorable for oxidation of the natural supply of the silt loam, or the sulfur in these soils, differing widely in their characteristics, is present in different forms. No doubt a considerable proportion of the sulfur in soils which, like the black clay, are distinguished by their high organic matter content, exists as organic combinations.

Peat soils generally contain considerable amounts of soluble sulfates of calcium and magnesium; consequently, the larger amounts of soluble sulfur obtained from the peat can not be regarded as an indication of conditions being much more favorable for the oxidation of its original sulfur content.

In all the mixtures where sulfur was included in the treatment, there has been a considerable production of sulfates, the amount of sulfur oxidized depending upon the soil and the treatment. Somewhat larger amounts of sulfates were extracted from the peat to which sulfur was added.

The quantities of sulfates found in the water extract of the acid silt loam are of interest as an index of the effect of the wide range of treatment, especially the different degrees of basicity provided, on the sulfur oxidation process. Although the amounts of sulfates resulting from oxidation of sulfur compounds naturally present in the acid silt loam are small as compared with the quantities found in the water extract of soils to which sulfur was added, they furnish an indication of the influence of differences in treatment on sulfofication.

Where sufficient calcium carbonate was present to satisfy the soil's requirement for base an increase of soluble sulfur was found. The addition of casein has also produced the same effect.

Considering in detail the results for the sulfur group of mixtures, including those with floats and casein in addition, it will be noted that different amounts of calcium carbonate have had an appreciable influence on the degree of sulfofication.

Where the treatment was sulfur, or sulfur and floats, the minimum oxidation of sulfur occurred when the total requirement of the soil for base was satisfied by calcium carbonate supplied at the rate of 4000 parts per million of soil. The sulfur as sulfates has tended gradually to increase with the lesser additions of calcium carbonate, being greatest where no basicity was supplied.

It will be noted that in the group of mixtures having casein with sulfur, the effect of the several amounts of calcium carbonate has been different. The indications furnished by the sulfur results when casein was included are that the optimum basicity for sulfofication was one-half the total lime requirement, the most soluble sulfur being found where 2000 pounds of calcium carbonate were added, rather than where no calcium carbonate was supplied, as was the case in the sulfur and the sulfur and floats groups of mixtures.

Rock phosphate, or floats, appears to have had a slight effect on sulfur oxidation when calcium carbonate was added. Comparing the sulfur results through the three applications of calcium carbonate in the mixtures receiving sulfur, and sulfur and floats, it will be observed that when floats were included the oxidation of sulfur has been slightly depressed.

ACIDITY AND ALKALINITY

From 50 to 80 per cent of the sulfur added having been oxidized, the acidity produced will have had an effect on other processes.

The figures for acidity stated as sulfuric acid show that it follows the order of the sulfur oxidized, gradually increasing with the smaller additions of base, and as would be expected, is greatest where no calcium carbonate was supplied to neutralize the acidity.

This relation between the maximum oxidation of sulfur and acidity exists only where casein was not included in the mixtures with sulfur. With casein and sulfur both added to the soil, the acidity where no calcium carbonate was present was decidedly less than that of corresponding mixtures without casein, the acidities of the sulfur mixtures being 511 and 666, while those of the mixtures which included casein with sulfur were 144 and 159 parts per million. This depression of the acidity of the water extract in the one case is evidently due to the influence of sulfur oxidation in limiting the transformation of proteid nitrogen so that it proceeded to ammonia only. Acidity being produced by the oxidation of sulfur, and no calcium carbonate being supplied, the basicity of the soil was not sufficient for the further change from ammoniacal to nitric nitrogen. Consequently, the increased accumulation of ammonia where casein was present has neutralized the acidity which has been produced through sulfofication.

While considerably more nitrogen as ammonia was found in the sulfur-casein mixtures than where casein was not included with sulfur and additions

of 2000 and 1000 parts per million of calcium carbonate were made, the acidity of the water extract has not been changed appreciably.

The alkalinity of the water extract, of course, was greatest where calcium carbonate was furnished to satisfy the total requirement of the soil, and nitrification and sulfofication proceeded less actively than when either casein or sulfur was present. It will be noted that this alkalinity has been decidedly decreased where the largest formation of nitrates from proteid nitrogen occurred, the amounts of nitric nitrogen being 400 and 440 parts per million. The calcium in solution as calcium nitrate was also greatly increased where the alkalinity was decreased.

The figures for alkalinity and soluble calcium, considered in relation to the maximum production of nitrates, strikingly illustrate the base requirement of the nitrifying process. Further indications of the effect of nitrification in depleting the soil's supply of bases is furnished by the increased acidity of the water extract of the soil mixture in which nitrogen supplied by casein has been partially changed to both ammoniacal and nitric forms in the absence of sufficient calcium carbonate for the more complete transformation to nitrates.

The solutions from the black clay were all alkaline. While the amount of sulfur oxidized in the clay soil mixtures was approximately the same as in the acid silt loam, the natural basicity of the clay was in excess of the amount of base attacked by acidity resulting from sulfofication. The water extracts from the peat were all acid regardless of sulfur oxidation.

NITRIFICATION

The chief purpose of the investigation which furnished the reported data has been to determine the effect of sulfofication on nitrification in acid and basic soils, and whether these processes have selective power to combine with specific soil constituents. While no attempt has been made to study the phenomena of nitrification and ammonification, the results have contributed some interesting information pertaining to the influence of different treatments on these processes.

The nitrogen data show that the transformation of proteid nitrogen in different types of soils is influenced by several factors. Considering first the nitrate nitrogen in the soils to which no nitrogen was added, it will be observed that the largest accumulation was in the acid peat; a smaller amount was found in the basic clay and the minimum production occurred in the acid silt loam. These figures parallel the total nitrogen content of the three soils regardless of their extreme variations in basicity and acidity. The nitrification process does not appear to have been greatly depressed in the decidedly acid peat. In this connection, mention is made of the fact that samples of acid peats taken during the summer months when conditions were favorable for nitrification showed a nitrate content in field soils of 200 parts per million.

Nitrification as an indicator of acidity or basicity of soils is quite strikingly

illustrated by the production of nitrates in the naturally basic black clay, as compared with that in the silt loam, which has a total requirement of approximately 4000 pounds of calcium carbonate per million pounds of soil.

The amounts of nitric nitrogen in the black clay mixture were practically the same whatever the treatment, while in the acid silt loam nitrification was decidedly increased when calcium carbonate was added. No mixtures of the black clay or peat included casein as part of the treatment on account of their natural supply of organic nitrogen; consequently, the comparison with respect to relations between basicity and nitrification is made between the silt loam receiving no casein and the other soils.

The maximum production of nitrate nitrogen in the acid silt loam occurred where calcium carbonate was added in amount sufficient to satisfy the soil's requirement for base. This accumulation of nitrate nitrogen equal to more than 80 per cent of the organic nitrogen added can not be regarded as the total quantity of the added nitrogen which has been changed during the whole period, since ammonification when casein or blood is the source of nitrogen may proceed more rapidly than the ammonia produced can be converted to nitrates, with the probability of there being a considerable loss of nitrogen.

The effect of increasing basicity is quite pronounced in the peat soil where no oxidation of added sulfur occurred. While the addition of calcium carbonate sufficient to supply the lime requirement of the peat soil has caused the largest production of nitrate nitrogen, this increase is only about double that in the peat receiving no calcium carbonate or other treatment.

The nitrogen results when sulfur has been oxidized in the silt loam mixtures receiving the smallest addition of calcium carbonate, one thousand parts per million, which is one-fourth the amount required to satisfy the lime requirement, show that this amount of base has very little effect on nitrification, since the amount of nitric nitrogen was practically the same as that recovered from the soil with its requirement entirely unsatisfied. This is so where casein was present as well as where the indications were obtained from the natural nitrogen supply of the soil.

Acidity developed by sulfofication has had a decided effect on the nitrogen transformation, especially in the acid silt loam where the greater variety of treatment including proteid nitrogen supplied by casein affords an opportunity for making more extended comparisons.

The nitrogen results for this soil have clearly shown that a measure of either the nitric or ammoniacal nitrogen independently would not have furnished correct indications of changes from proteid to other forms of nitrogen when oxidation of added sulfur has occurred, for the reason that with certain conditions of treatment no appreciable amounts of nitrates were produced, the transition being to ammonia only, while under other conditions the change has proceeded more completely and practically all of the nitrogen was present as nitrates.

The transformation from ammonia to nitrates is depressed in the silt loam when sulfur has been oxidized, unless the acidity produced has been neutralized by calcium carbonate. This was also true for the nitrates in the acid peat. Sulfofication has had very little influence on nitrification in the basic clay, the quantities of nitrates found being practically the same regardless of sulfur oxidation.

One of the interesting points brought out by the work on nitrogen is the quantitative relationship between the results for nitrates and the calcium carbonate additions to the silt loam where the indications were obtained from the natural nitrogen supply as well as where proteid nitrogen was supplied by casein.

When sufficient calcium carbonate was present to satisfy the soil's requirement, excluding that necessary for the neutralization of acidity developed by sulfofication, the maximum quantities of nitrates were produced, the quantity found gradually decreasing with the smaller additions of carbonate and the ammoniacal nitrogen increasing. Although calcium carbonate in amount considered to be sufficient to satisfy the soil's demand for base exclusive of the additional requirement resulting from the oxidation of sulfur, increased the production of nitrate nitrogen where sulfur and casein were included together this quantity of calcium carbonate, however, was not adequate to satisfy the requirement of the soil for base and at the same time neutralize the acidity produced by the oxidation of sulfur. Evidence of this is furnished by the amounts of nitrates recovered from the mixtures which included casein with and without sulfur, and additions of calcium carbonate made at the rate of four thousand pounds per million. Where no sulfur was added, the quantity of nitrates recovered was 440 parts per million and where sulfur was oxidized there was a decrease to 295 parts per million.

AMMONIFICATION

When the results for nitrogen as ammonia are studied in relation to the nitric nitrogen figures obtained under the conditions of this experiment, they are of special significance in that they show the influence of various factors on the transition from the one form to the other.

Excluding the mixtures of peat soil, the quantities of ammonical nitrogen found are relatively small as compared with the ammonia extracted from the soil mixtures which included casein and sulfur. Oxidation of sulfur in the naturally basic clay soil has produced no variation in the amounts of ammonia, the slight differences exhibited by the results being well within the limit of analytical error. This uniformity conforms with the regularity observed between the results for nitrates and ammonia throughout the acid silt loam group of mixtures, that is, when conditions were made favorable for the production of nitrates by supplying base, the amounts of ammonia were small.

One relation between the two forms of nitrogen which is shown by the results is that the amounts of ammoniacal nitrogen vary inversely with the nitrates.

Casein being included in the silt loam mixtures and provision made for different degrees of basicity, the results for these mixtures are of interest when considered in detail.

It will be noted by referring to the data included in table 1, that the maximum accumulation of ammonia in the sulfur-casein treated mixtures receiving 4000, 2000 and 1000 pounds of calcium carbonate per million, is in direct relation to the largest amount of sulfur oxidized. Nitrogen as ammonia and sulfur as sulfate in these groups of mixtures were recovered in largest amount where calcium carbonate was added at the rate of two thousand parts per million of soil.

Where acidity was produced from sulfofication and no calcium carbonate was available to neutralize it, the transition from proteid to other forms of nitrogen proceeded to a slight extent only beyond ammonia, as the conditions were not favorable for the further change to nitrates and only small amounts of nitrogen in this form were found.

The results for ammoniacal nitrogen in these mixtures compared with the figures for acidity furnish evidence that in the absence of calcium carbonate, the ammonia produced neutralizing the sulfuric acid formed was combined as ammonium sulfate, since the acidity of the water extract was decidedly less than was found for similar mixtures where ammonifying organisms did not have a supply of proteid nitrogen to transform into ammonia. However, the acidity figures when basicity conditions were apparently optimum for sulfofication do not indicate that the ammonia formed in largest amounts from casein has materially changed the acidity from that where no casein was included in the mixtures with sulfur. The increased quantity of soluble sulfates associated with the ammonia present where two thousand parts per million of calcium carbonate were provided may be partly ammonium sulfate.

In these cases where conditions resulting from sulfofication, and the presence of insufficient base were such that the transformation from ammonia to nitric nitrogen was largely prevented, there would undoubtedly be a fixation of a part of the ammonium sulfate formed, since it is an established fact that soils have the capacity for fixing considerable amounts of ammonia from added ammonium salts. It would seem that following the fixation of ammonia from ammonium sulfate formed, whether this phenomenon of absorption or fixation is the result of chemical interchange or due to physical or other causes, there would be a consumption of calcium, magnesium or other base by the sulfate radicle. Since ammonium sulfate is regarded as a physiologically acid salt, in field soils the possible utilization of ammoniacal nitrogen by growing plants would be a cause contributing further to the depletion of soil bases.

Whether the ammonia is changed from its combination as sulfate directly to nitrous acid and nitrate, or whether there is an intermediate reaction, a base will be required, as is evidenced by the results for ammoniacal and nitrate nitrogen in the soils where sulfur was oxidized and variations in basicity existed.

In this connection, two questions are suggested. To what extent will tricalcium phosphate serve as a source of basic calcium in soil which, while not supplied with calcium carbonate, contains minerals which may be attacked by acidity developed through nitrogen transformations, or by the sulfate radicle of ammonium sulfate? The acid radicle will be a factor regardless of whether ammonia is oxidized by nitrite bacteria, fixed by soil or utilized in part directly by growing plants.

And further, if the soil is supplied with calcium carbonate, will the nitrous acid exercise any selective action and unite with the calcium of tricalcium phosphate in preference to calcium present as carbonate?

Without the data for calcium and phosphorus which will be obtained as a part of the investigation, at present not completed, the nitrogen results must be regarded as indicative only. However, the figures for nitric nitrogen found in these mixtures of the acid silt loam soil where floats were included and either one thousand parts of calcium carbonate or none were added, furnish very slight evidence of the calcium in tricalcium phosphate mixed with the soil at the rate of one thousand parts per million having functioned as a base.

When no change was brought about in basicity by calcium carbonate or in acidity by the oxidation of added sulfur, a part of the proteid nitrogen furnished by casein has been completely changed to the nitrate form and a part to ammonia only. The ammonia found was considerably less than was present in mixtures which were similar except that added sulfur was oxidized, and the nitrates were half the amount produced when the base requirement of the soil was satisfied.

SUMMARY

Oxidation of elementary sulfur in soils devoid of base depresses activities of nitrifying organism.

Although basicity was supplied by calcium carbonate in excess of the requirement of the soil, the yield of nitric nitrogen from casein was considerably decreased by oxidation of added sulfur.

A further decided depression occurred with decreased basicity, so that a quantitative relationship exists between the yields of nitrates and calcium carbonate additions.

The increasing amounts of ammonia which accompanied the decreased yields of nitrates when sulfur was oxidized, can not be considered as indicative of sulfofication having exerted a stimulating effect on ammonification.

This inverse relation between ammonia and nitrates must be interpreted as existing rather because the deficiency of base necessary for neutralizing sulfuric acid has inhibited the transition from ammonia to nitric nitrogen. The ammonia formed neutralized the acidity and remained as ammonium sulfate.

REFERENCES

(1) ALLEN, E. R. 1915 The determination of nitric nitrogen. *In* Jour. Indus. Engin. Chem. v. 7, p. 521.

THE IMPORTANCE OF MALARIA TO AGRICULTURE AND STUDIES ON MALARIAL SOIL

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It is well known to all, whether it be in Africa or America, how very often malaria is an obstacle to the permanent occupation of the soil by man. As a matter of fact, there are numerous cases of expeditions of explorers and travelers destroyed, and of attempts at colonization impeded on the "Dark Continent." It is also well known that in the cutting of the Panama Canal the major obstacle was malaria in conjunction with yellow fever.

But even in Europe malaria is still an obstacle to the progress of agriculture. Southern Italy, southern Spain, southern Russia, Greece and nearly all of the Balkan Peninsula are strongly malarial. As to farming in these regions, as the saying is in Italy, internal colonization finds its principal obstacle in malaria. These regions are characterized:

(a) By lack and scarcity of rural population. On the other hand, population is congested in the settled centers. Very often these centers are very distant from each other.

(b) By the predominance of large estates under extensive culture as compared with small estates under intensive culture.

(c) By lack of country houses and roads.

(d) By disorders of stream action (erosion) due to lack of systematic handling of the land, which is manifested by the torrents in the deforested hills with inundations and swamps in the low lands.

(e) Finally, by the prevalence of malaria, which above all else in the summer interferes with the housing of workers in the country.

Besides this group of causes, which in their turn are effects, historians point out remote causes in the large estates of the Roman Empire, in the Saracenic invasions (which compelled the population to take refuge in the hills) in feudalism and in feudal legislations, in brigandage, in the occupations by strangers (enemy or conquerors), in the destruction of forests, etc. These conditions, until a short time ago, formed a closed cycle which nothing could break; extensive culture, the large estate, the swamp-engendered malaria, which in turn gave rise to the depopulation. This depopulation resulted in extensive culture, large estates, pasture feeding, and the abandonment of the soil.

However, the studies of Laveran, Celli, Marchiafava, Ross, Manson and Grassi, had made certain that malaria is propagated exclusively by means of mosquitoes of the genus *Anopheles*. They demonstrated that the protozoan parasite passes a closed evolutionary cycle between the *Anopheles* and man. It was thought for a certain time that by improving the health of the malarial person or curing with a specific remedy, and also through keeping healthy people from contracting the disease from bites of infected *Anopheles* by screening, malaria would be destroyed in a few years. Thus the principal obstacle to the possession of much land would be removed.

It was at this time that the malarial flagellum was considered a conquered giant. The illusion faded very quickly. The curing of malarial patients in the preepidemic period was proved to be too inefficient in diminishing the quantity of malarial virus quickly. Mechanical protection with netting on doors and windows, and with gloves and masks, appeared to be a form of nonsense practical for the most part only as a luxury.

In the meantime, however, a new horizon had formed about hygiene. It was said that, since malaria is carried by the *Anopheles* which has its home in water, removing the water from the malarial places by means of well-conducted improvements, ought to effect the reclamation of the malarial locality by removing the means of malarial propagation.

It had to be recognized very soon that the mere presence of *Anopheles* was not the true criterion of the possible prevalence of malaria. The number of *Anopheles* was not synonymous with the epidemic; that is, it was possible to have entire regions badly infested, with little malaria prevalent, and on the other hand, it was possible to have a severe malarial condition with a ridiculously small number of *Anopheles*. Thus it was discovered that there could be freedom from malaria with swampy conditions and a prevalence of *Anopheles*. It was ascertained that there are zones in all latitudes in which malaria arises, which are notoriously swampy and give rise to a very abundant *Anopheline* fauna, but that malaria in these places is well-nigh only a matter of historic record.

This was enough to demonstrate that the equation

$$Man \times Anopheles = Malaria$$

was incomplete and that instead it should be expressed

$$Man \times Anopheles \times x = Malaria$$

In continuing researches on the aquatic habitat of the *Anopheles*, its enormous facility for adaptation was recognized. It was seen that the phenomenon of *Anophelism* without malaria was the rule and not the exception. Hence the etiological formula, that where the *Anopheles* exists there is malaria, should be changed into the following which the author has proposed: Where there is malaria there is the *Anopheles* which transmits it.

But whatever other disillusions these last proofs did not bring forth, there was a true debacle of every scientific application to practice!

It was thus feared for a while that we should have to renounce every hygienic effect of drainage. It was thought that it was mere nonsense to hope that drainage could be so well conducted as to remove the possibility of propagating *Anopheles* in a territory in which they had been established for centuries. Since the *Anopheles* might not find any more water truly stagnated, they would content themselves with wells, drinking fountains, ponds in gardens, wash tubs and puddles, and in the absence of fresh waters, would end up in making use of salt waters.

It would have been a case of despairing of every progress based on the new discoveries, if two new proofs had not come to our aid. These can be summarized thus:

(a) It is possible to live in malarial places by means of modern prophylaxis, both general and quininic.

(b) Drainage improvement, which has behind it agrcultural improvement, that is, intensive cultivation of soil already malarial, makes the malarial regions sanitary again.

In the first point there is a plebiscite of scientists and conscientious experimenters. As to numerical examples, they do not stand in any need. Those reported in the table of Celli were published in the paper on malaria, entitled "Work of the Society for the Study of Malaria from 1898 to 1908." On the second point, there is also a plebiscite determined by a series of objective and systematic studies.

In fact, from studying the major part of Italian improvement (drainage), it has been concluded that where for particular reasons it has been possible to establish agricultural improvement, immediately after drainage improvement there has also been a hygienic improvement. The contrary resulted where it was not possible to have agricultural improvement as a complement to the drainage.

The author's station has carried no light contribution of studies in this sense. Among these may be recorded:

(a) Studies on the improvement of the province of Reggio Emilia (northern Italy) which have made the fact prominent that the said province can be divided into two regions: a low region with much surface waters, with a great prevalence of *Anopheles* and a small amount of malaria; and a high region without surface waters worthy of note, with a prevalence of *Anopheles* sufficiently serious, yet in which it may be claimed that malaria has disappeared. The only historic difference between the two regions lies in the fact that in the high part of the Reggian Plain, drainage improvement is antiquated while on the low region it is relatively recent.

(b) Observations made about the province of Macerata (one of the five provinces of Italy immune from malaria for some time and under intensive agriculture), which led to establishment of the fact that while malaria had not existed at the place for about one century past, with the last drainage improve-

ment and in the latest rice fields, the prevalence of *Anopheles* is still very great and has an influence upon homes near the city.

(c) Studies and researches upon the celebrated ex-Lake Fucino, drained by the House of Torlona. These show that if in the Fucine Region the swamp, has always existed, and hence presumably the *Anopheles* also, malaria there has never become intensified. There was an exception to this in a period of a few years which coincided with the work of drainage and which gradually disappeared while agricultural improvement was becoming established on the drained banks.

(d) Observations and verifications in which we are assisting in many localities of the south. If in itself malaria has very often impeded the advance of agricultural improvement in its intensity, the fruitfulness of the climate has also permitted of other means of resanitation following drainage and agricultural improvement. This has taken place in lands of pronounced malarial conditions. It is noteworthy from this viewpoint, in the negative sense, as regards portions of Vico in the drainage of the smaller basin of the Volturno, until almost up to the reign of Ferdinadn II.

The Bourbonic administration had well understood that to complete the gigantic works accomplished, it was necessary to add private to the public improvement. In addition to this, in so far as the administration was able, it had allotted to the inhabitants of Vico di Pantano a good portion of the reclaimed land, constructing for the most part a small house for each portion. The air was still "mefitic," to use the language of the time. This did not permit the peasants to sojourn in the open country. The houses were abandoned (the remains may still be seen) and the portions left were reformed into the large estate.

The Colony of Battipaglia, constructed almost contemporaneously as a place for fugitives from the earthquake of Melfi, also failed in its scope. There was even a greater failure also about the plain of Eboli, as every inhabited center made its influence felt for a considerable radius about itself. On the other hand, the gardens of Schito and all the valley of the Sarno succeeded in becoming sanitary again in a little less than half a century. Here the dominant factor was the most intensive of culture, as is followed in turning the soil four times annually in cultivating kitchen gardens. It is all the more worth while to consider the swamps of Naples, from which malaria has disappeared (or nearly so) since each zone could be cultivated.

Nor is there a lack of very recent examples. Let the reclamation of the estate of Pantanaat Castelnuovo Vallo suffice for all. Here the disappearance of the epidemic has kept pace with an increase in farming and with the breaking up of the large estate, thanks to quininic prophylaxis.

(e) Studies and observations (not yet edited) made on the swamps in the provinces of Pisa, Livorno and Lucca, which are almost entirely without malaria. Studies on Massarosa, Massachiucoli and the vicinity of Lucca, etc., have shown that here also the only explanation of the fact is that not one

square meter of soil not submerged is left uncultivated. Even the water, so to speak, is cultivated, being used in the rice fields.

(f) Studies and observations (not yet edited) made in France, in the vicinity of Paris, Sarthe (Le Mans, Bonnetable), and Normandy (Alengon, Le Ferté Bernardete) which have confirmed that these regions, in which malaria is not even more than a matter of historic record, abound in swamps and in *Anopheles*. However, they are under intensive agriculture in all regards.

In other words:

(a) A positive connection certainly exists between intensive cultivation of the soil and a diminution and even a disappearance of malaria. This is not necessarily in conjunction with a decrease in surface waters, capable of harboring *Anopheles* larvæ.

(b) Neither the numerical gravity of malaria (or the coefficient of morbidity) nor the gravity of the infections (or the coefficient of perniciousness) is as a matter of fact in relation to the number of *Anopheles* of a certain territory.

(c) It has been impossible as yet to find any difference between the innocuous *Anopheles* of a reclaimed zone and the harmful *Anopheles* of a pestiferous zone.

(d) In the major number of cases it has been and is entirely possible to see that the spontaneous retrocession of malaria has been contemporaneous with the advance in the intensification of culture (or of the resumption of farming operations). This intensification was not necessarily for the entire territory so as to give the idea that it was due to a decrease in surface water. It was done on the parts free from water and therefore susceptible to culture. From all this the only conclusion in keeping with logic is that the remanagement of the soil has an unfavorable influence on the cycle *mosquito—man—mosquito* to the point of breaking its implacable continuity. This can be practically expressed by saying that the *Anopheles* of the cultivated zones become incapable of transmitting malaria. This influence must be specific even in spite of the fact that rare exceptions to this rule have been indicated. This may mean that the specific condition is not always present.

It may also be that such a condition may exist as the presence of a germ in the ground in a stage unknown to us, and which is killed by conditions of environment created by cultivation. We cannot claim to know all that may be known some day on the nature of protozoa. We should be constantly on our guard against asserting that we have added the pillars of Hercules to knowledge in any field of biology. From what we continually apprehend from the best-known germs (or those we believe to be best known) nothing authorizes us to admit it, because many other hypotheses can be made which have all the more reason to be, but are invalid, which are equal to the first from every basis of experimentation or observation.

It is very true that with this hypothesis of the malarial germ in the soil, the autoctonal cases of malaria (occurring in 8-day periods) also would be explained, which not even A. Celli denies. Even the hypothesis of a hyperparasite which was favored by the cultivation of the soil and which ended with

the destruction of the amatozoan in the *Anopheles*, the hypothesis of a relative immunity (absolute immunity is excluded) introduced into the *Anopheles* with the change of vegetative nutriment offered them by cultivation (analogous to the softening effect on plants from fertilization and cultural practices) and many other hypotheses have an equal right to be offered. Each of these is as worthy of consideration as any of the others; and who knows that they are not all true and that they are not explanations, for the present more or less plausible, of the fact which no one can deny, namely, that if the soil is put under management again, malarial epidemy becomes attenuated and disappears?

There is a tendency to reduce every thing to formula. The equation

Malarial Man × *Anopheles* = *Malarial Epidemic*

as has been said, no longer stands. The other by A. Celli

Malarial Man × *Anopheles* × *xyz* = *Malarial Epidemic*

is nothing but the expression of our ignorance, because no one has given us the value of the physical, biological and social factors which are represented by the unknown *xyz*. The formula proposed by the author: *Where there is malaria there is the Anopheles that transmits it*, is also destined to restate the bearings of the old experience which people had with malaria, a disease essentially autoctonal and allied with the soil. This may bear relation to the true, perhaps indestructible, bearings of modern science, according to which malarial epidemy is maintained in man only with the bite of the *Anopheles*.

Moreover, science must explain exhaustively the mysterious connection which passes between soil and malaria, or find the true connecting links between the exclusively and closely parasitarian theory and the telluric theory of Tommaso Crudeli.

It is convenient to observe that, seduced by the bewildering exclusiveness of the doctrine of the malarial mosquito, we have disguised the studies of the soil as a malarigenic factor. It is still necessary to turn to the soil, which in its bosom holds the waters that harbor the *Anopheles* larvæ. This soil in conjunction with the waters, forms the mud which often harbors and permits the eggs to hatch. Plain looking waters sustain the aquatic plants of the most various small forms. It has a part in determining the plankton of swampy waters and is always connected with the micro- and macro-fauna, with which *Anopheline* generations come continually into biological conflict. In brief, it permits the natural and artificial development of the plants from which 90 per cent of the *Anopheles*, which are essentially phytophagous, get their nutriment.

Still no fact is available to furnish conclusive proof. The culture of malarial parasites is yet to come, notwithstanding the attempts of Bass and Johns. The study of swamp plants has possibly brought in only a ray of light. Celso Ulpiani's report on the relation between colloids of the soil and malaria has introduced a pleasing intimation.

He says:

The formation of swamps is, according to me, a colloidal phenomenon, as is also malaria, which in our warm climates is always found in conjunction with the existence of swamps. This includes a colloidal problem. I do not know what connection may exist between malaria and swamps, but a connection must exist. The biological cycle of the fatal plasmodium must in some manner be connected with the permanence of the hydrozoa of the soil and with the high temperatures of the climate. Colloidal chemistry will certainly give the explanation for the indissoluble relation which exists among swamp malaria and warm climate. When floods occur in the bottom of a valley in our climates and the assiduous and tenacious farmer does not break up the colloidal deposits resulting left by floods with the plow, the spade and hoe, it favors the agglomeration of these colloids. If for years and years agriculture becomes absent, the colloids deposited on the washed and leached soil begin to obstruct the pores of the subsoil. They glue the particles together while with the diminished access of air the organic matter becomes humified. This protects the organic colloids more and more from coagulation. Upon this soil, which, so to speak, may be said to be "varnished," and which no longer takes up the water, the swamp is found with its black muddy waters. Its mud is slippery and insidious and its flora useless. It has its mosquitoes, its wandering lean animals and a sparse human population degraded by fever.

This is purely a pleasant conception, but one which deserves to be taken into serious consideration by studious people.

Hence the author believes that the courses to be followed by the studious must be new and diverse in the study of the problem. It is true that problems of this nature are hardly solved in a direct way. For the most part, their initial solution (or the first clue) is in accordance with the case. But it is also true that the case generally helps those who place themselves in a position to be helped by it. It is therefore necessary to study malarial soil in all its manifestations in contrast to the now malarial soil, using both fundamental systems of scientific study—observation and the experiment. Observation should include retrospective observation, that is, the history of the soil, either geological history or that concerned with its recent vicissitudes, in other words, the history of its agriculture. It is true that in recent times there has been an outcropping of historical studies on malaria, to which A. Celli has given impetus. For years he collected precious material for the historic study of Roman malaria. He also gave an accurate and perhaps complete bibliography.

The studies which we owe to Ronald Ross, in collaboration with C. C. Ellett, are entitled "Malaria, a Neglected Factor in the History of Greece and Rome." There is also the one by Paul Roux, "The Agricultural Question in Italy." Studies by Dr. E. Genovese on Calabrian malaria, "The Roman Estate," are also worthy of note, and perhaps those made by this station concerning the places improved, which have been reviewed, are not to be neglected. Even in other times the historic study of malaria was overshadowed. Memoirs contained in the first and only volume on the "Annals of Improvements," published through the care of John Savarese, are entitled to honor. The same may be said above all of that by the Canon, John Scherillo, entitled "On the Air of Baia at the Time of the Romans."

But chiefly, the most neglected reports in these studies are of course those interceding between malaria and agriculture, not only that which reflects

"rural economy," but also, and above all, "agricultural technique." The study of the latter is notoriously difficult and actually always incomplete. If, as a matter of fact, we know little about Roman agriculture, notwithstanding the works of Cato, Varro, Virgil, Pliny, Columella, etc., we know less about Grecian agriculture. We know almost nothing about Mediæval agriculture, wherein ever the sources to draw on are lacking. Books and the history of modern agriculture also are lacking.

And yet it may be absolutely necessary to be familiar with this history as to "agricultural ground and malaria" in order to obtain information therefrom in our day on the manner of conducting agricultural improvements. It tells us how much to expect and to count on with regard to the above in order to arrive at the definitive hygienic resanitation against malarial plagues. As to experimentation, it can be and should be carried out in various ways, and, moreover, with more than the ordinary laboratory researches; more properly,

(a) By setting up experiments *ad hoc* in which it shall be possible to reproduce the natural conditions exactly.

(b) With experimental demonstration camps true and proper.

(c) With adequate collections, in which the studious person with his talent shall be able to find as much about the subject as the research of the moment warrants.

The station, under the author's direction, which has been occupied with the study of reports on malaria and agriculture for 17 years, is conscious of the very numerous reports which exist but are not known in the field just mentioned. It is conscious of the difficulties and the relative necessity of their solution. It believed it to be well to confront the problems directly, studying also the swamp plants, the plankton of the swamps, the reports of the *Edaphon* with the problem on malarial soils in connection with the colloidal state and the prevalence of malaria in the territory. For this purpose a collection of material has been created necessary according to the consideration which we have mentioned. In fact, the station has a number of enactments for the production of artificial swamps more or less extensive, ground held under open pasture and pastures with rhyzomacious plants, ponds with water-proof bottoms, both elevated and sunken, also ponds with water-proof sides and porous bottoms, an experimental aquarium, etc. In addition to this, it is providing itself with special equipment without forgetting the proper collections, and above all, collections of soil from swampy and malarial places. It is providing itself with special products of the swamps, a small collection but very interesting, and destined to complete the studies on the *Edaphon* and the plankton in conjunction with malaria. It is the author's intention to publish a catalog which is now in the course of compilation. This is for the use of persons making studies in this field, with the purpose of increasing collections by means of exchanges and communications.

All the collections mentioned are catalogued. The author hopes that at the end of the war he may be permitted to place his equipment and collection at the

disposal of investigators who may wish to collaborate with him for the extinction of one of the most burning plagues of all humanity, the dreaded malarial infection.

BIBLIOGRAPHY

(1) Di Pace, I. 1916 A proposito della malaria da sterri. Problemi di malariologia. *In* Malariologia, v. 9, 1916, p. 23; 57; 80; 103; 134; 171.

(2) Guarnieri, G. 1908 Il versante Jonico della Basilicata. Ricerche sulla sua malaria e sulle bonifiche ivi in esecuzione. *In* Atti Soc. Studi Malaria, v. 9, p. 351–379.

(3) Guarnieri, G. 1908 La bonifica del Golfo di S. Eufemia. *In* Atti Soc. Studi Malaria, v. 9, p. 45–417.

(4) Guarnieri, G. 1909 Il passato a il presente dell'Agro Telesino rispetto al suo bonificamento. Nota I. Benevento, Tip. D'Alessandro.

(5) Guarnieri, G. 1909 La malaria nel Bacino del fiume Diri in Provincia di Caserta. La piana di Cassino. Extract from Relazione del Comitato Agricoltura Antimalarico di Terra di Lavoro per l'Anno 1908 a S. E. il Ministro d' Agricoltura, Caserta, Turi e Figli.

(6) Rossi, G. 1902 Dell'influenza di alcune bonifiche sulla malaria in provincia di Napoli e Terra di Lavoro. *In* Atti Soc. Studi Malaria, v. 4, p. 377–398.

(7) Rossi, G. 1903 La bonifica delle paludi di Napoli dal punto di vista della malaria. *In* Il Benessere, n. s., v. 10, Feb. 1903, fasc. 8, p. 225–230.

(8) Rossi, G. 1904 Sulle vicende agricule ed igieniche della Piana di Fondi e Monte S. Biagio in rapporto colle bonifiche ivi eseguite. *In* Ann. R. Scuola Sup. Agr. Portici, ser. 2, v. 6, 30 p.

(9) Rossi, G. 1904 Malaria e bonifiche della Piana di Fondi e Monte S. Biagio. *In* Atti Soc. Studi Malaria, v. 5, p. 653–662.

(10) Rossi, G. 1905 Malaria e bonifiche del Bacino inferiore del Sele. *In* Atti Soc. Studi Malaria, v. 6, p. 397–410.

(11) Rossi, G. 1906 Le bonifiche delle provincie di Napoli, Caserta Salerno, nei loro rapporti colla malaria e coll'agricoltura. *In* Portici, R. Scuola Superiore di Agricoltura. La R. Scuola Superiore d'Agricoltura in Portici nel Passato e nel Presente. 1872–1906. Portici, p. 168–170.

(12) Rossi, G. 1907 Sulla bonifica idraulica, agraria ed igienica della Valle del Crati in Provincia di Cosenza e sui lavori giá in essa eseguiti. *In* Atti Soc. Studi Malaria, v. 8, p. 623–638.

(13) Rossi, G. 1907 Malaria e bonifiche della Provincia di Reggio Emilia. *In* Atti Soc. Studi Malaria, v. 8, p. 571–593.

(14) Rossi, G. 1908 La bonifica del lago Salpi nei suoi rapporti colla malaria. *In* Atti Soc. Studi Malaria, v. 9, p. 381–404.

(15) Rossi, G. 1908 La bonifica della Valle dell'Idro in provincia di Lecce ed il suo significato per l'avvenire del territorio Otrantino. *In* Atti Soc. Studi Malaria, v. 9, p. 419–427.

(16) Rossi, G. 1909 Malaria e bonifiche della Provincia di Reggio Emilia. Nota II. Consorzi di difesa delle sponde della Secchia. *In* Atti Soc. Studi Malaria, 1910, v. 11, p. 331–340.

(17) Rossi, G. 1909 La bonificia agraria del Mezzogiorno d'Italia nei suoi rapporti colla bonifica idraulica ed igienica. Relazione al II Congresso agrario. *In* Atti del I Congresso Agrario Meridionale. Napoli, Ferrante, 1909. *In* Propaganda Antimalarica, no. 6, 1909. Napoli-Tip. Tocco e Salvietti. *In* Atti Soc. Studi Malaria, v. 10, p. 483–499. 1910. *In* Il Clinino dello Stato delle sue Origini ad Oggi. Il Ministero delle Finanze, Direzione Generale delle Privative. 1910.

(18) Rossi, G. 1906 Malaria e bonifiche dei "Campi Flegrei." Ann. R. Scuola Sup. Agr. Portici, ser. 2, v. 6, 20 p.

(19) Rossi, G. 1910 Necessità dello studio storico dei rapporti fra malaria ed agricoltura. *In* Atti Soc. Prog. Sci. 4th reunion, Napoli, December, 1910. Roma, Tip. Bertero, 1911. p. 856–858.

(20) Rossi, G. 1911 L'ex lago Fucino ed il paludismo senza malaria. *In* Atti Soc. Studi Malaria, v. 12, p. 287–313.

(21) Rossi, G. 1912 Studi e ricerche sull ex lago Fucino. Contributo allo studio del Paludismo ed Anofelismo senza malaria. *In* Ann. R. Scuola Sup. Agr. Portici, ser. 2, v. 11, 31 p.

(22) Rossi, G. 1911 Paludismo ed anofelismo nella Provincia di Marcerata. Nota preliminare. *In* Ann. R. Scuola Sup. Agr. Portici, ser. 2, v. 10, 9 p.

(23) Rossi, G. 1913 Malaria e bonifica della Piana di Catania. Studi e ricerche. *In* Ann. R. Scuola Sup. Agr. Portici, ser. 2, v. 11, 23 p.

(24) Rossi, G. 1913 Sul bonificamento definitivo della Piana di Fondi e Monte S. Biagio. Studi e ricerche. *In* Ann. R. Scuola Sup. Agr. Portici, ser. 2, v. 11, 20 p.

(25) Rossi, G. 1914 Agricoltura e malaria della Valle dell'Enza. Studi e ricerche. *In* Ann. R. Scuola Sup. Agr. Portici, ser. 2, v. 12, p. 391–418.

(26) Rossi, G. 1914 Sulla dottrina zanzaro-malarica. *In* La Propaganda antimalarica (Rivista di Malariologia) anno 7, no. 3, Napoli, Tip. Tocco, p. 78–82.

(27) Rossi, G. 1915 Relazione su alcune prove culturali eseguite nell'ambito della colmata di Licola. Verbal extract no. 59 from the meeting of October 17, 1914, of the second session of the Commissione Centrale per le Sistemazioni Idraulico-Forestali e per le Bonifiche. Roma, Tip. del Genio Civile.

(28) Rossi, G. 1915 Sulla coltivazione dei futuri terreni della campagna vicana. *In* Ann. R. Scuola Sup. Agr. Portici, v. 13.

(29) Rossi, G. 1916 Il rimboschimento delle due nei riguardi igienici anche in rapporto colle bonifiche idrauliche. Relazione al III Congresso Forestale e I Congresso per l'Irrigazione e Discussioni Relative. *In* Atti. Soc. Studi Malaria. Portici.

(30) Rossi, G. 1917 I nuovi orizzonti delle ricerche antimalariche ed i nostri impianti sperimentali. *In* Malariologia, v. 10, p. 73–83.

(31) Rossi, G., D'Elia, E., and Campbell, C. 1914 Il disseccamento definitivo della Piana di Fondi e Monte S. Biagio nei riguardi idraulici, agrarie di gienici. Portici, *In* Ann. R. Scuola Sup. Agr., ser. 2, v. 12, p. 617–653.

(32) Rossi, G., and Guarnieri, G. 1906 La bonifica del Vallo di Diano nei suoi rapporti colla malaria. *In* Atti Soc. Studi Malaria, v. 7, p. 387–402.

(33) Rossi, G., and Guarnieri, G. 1906 Palude, malaria e bonifiche dell' Agro Sarnese. *In* Atti Soc. Studi Malaria, v. 7, p. 403–414.

THE INVERSION OF CANE SUGAR BY SOILS AND ALLIED SUBSTANCES AND THE NATURE OF SOIL ACIDITY

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By the earlier investigators the inversion of cane sugar was believed to be catalyzed by hydrogen ions and by no other non-organized substance. (Inversion by enzymes will not be considered in this paper.) Ostwald (67), Arrhenius (9), Palmaer (69), Ley (52), and others believed so implicitly that inversion could not be produced in any other way that, whenever discrepancies appeared in their results, they were attributed to side reactions taking place, or to physical disturbances in the system.[1] Thus were hydrogen ions detected, and hydrogen-ion concentration determined—by measuring the rate of inversion of sucrose (95). The majority of chemists today believe as did those earlier workers, though some would modify the hypothesis to include undissociated hydrogen as also having catalytic action in some cases.[2]

From time to time phenomena have been recorded which at first sight appeared to be instances of inversion where acid hydrogen was not present. But in these cases it was never conclusively proven that acid hydrogen was not present either at the start, or formed later through the decomposition of the sugar itself. Cane sugar is quite easily decomposed, and among the products of decomposition, substances of an acid character seem to predominate. Thus often conditions may be such that although no acid is present at the beginning of an experiment, it may later be formed, inversion then taking place (10, 99). Such cases are recognized by determining at intervals the amount of sugar inverted, when it is found that as time goes on, the sugar is inverted at a greater and greater rate. On the other hand, if the action were due only to acid present at the beginning, then the rate of inversion must fall off as the reaction proceeds, since according to the mass law, the amount of sugar acted upon at a given time must depend upon the amount present at that time to be acted upon. Rayman and Sulc and others (49, 76, 85) have found that pure water causes inversion at high temperatures; the rate is low at first but as the heating continues it becomes greater. The initial inversion is attributed to that slight quantity of hydrogen ion always present in water, and the increase in rate, to acid produced through the decomposition of the sugar.

[1] Hence arose the ideas of "active" and "inactive" sugar molecules, stimulation of the catalytic activity of hydrogen ions, increased rate of movement of hydrogen ions, internal friction in solutions, increase in the osmotic pressure of the sugar in solution, and so on.

[2] Acree (4) and Snethlage (86) believe that in the case of the stronger acids undissociated acid hydrogen is even more active in causing inversion of sugar than hydrogen ion. See also Lamble and Lewis (50) and Harned (39).

Inversion by metals and metallic powders has been observed by Rayman and Sulc (76), Lindet (53), and Plzak and Husek (73), but again we must conclude that the sugar is first decomposed, and it is the products of this decomposition that are effective (99). Inversion by sunlight (75), the electric current (66), ultraviolet light (15, 29), and radium (46) are to be explained in the same way.

The action of salts on sucrose has been carefully studied. As should be expected, if a salt hydrolyses in solution with the formation of acid hydrogen, then inversion must take place. Such has been found to be the case with cadmium, zinc, lead and aluminum nitrates (101), manganese, zinc, iron and aluminum sulfates, iron, manganese, lead, cadmium, and mercuric chlorides (57), chromium sulfate (103), aluminum and copper chlorides, cadmium, copper and nickel sulfates (45). Salts of bases with acids of equal strength, or of strong bases with weak acids, do not invert.[3]

Geerligs (33) made the interesting observation that while invert sugar exhibited no action on sucrose when alone and neither did such neutral salts as the chlorides of potassium, sodium, lithium, calcium, strontium, barium and magnesium, yet if invert sugar was present in solution together with any of these salts, hydrolysis of the sucrose did take place. It was explained by postulating that through hydrolysis of the salt, and a combination of the basic part with the monose sugar, the acid was left free to exert the characteristic inverting effect. The following equation was given to explain this:

$$\underset{\text{salt}}{MA} + \underset{\text{water}}{HOH} + \underset{\text{glucose}}{C_6H_{12}O_6} = \underset{\text{basic glucose compound}}{MOH\ C_6H_{12}O_6} + \underset{\text{ionized acid}}{HA}$$

Contradictory results, however, were obtained by Deerr (28).

The literature on the subject of sucrose inversion is voluminous, but it has never been proven that this reaction can be effected in any other chemical way than through the presence of acid.

INVERSION OF CANE SUGAR BY SOILS

That soils will cause inversion of cane sugar has already been shown by Tacke and Süchting (94), and by Hanley (37). It seemed to these investigators that organic matter was the principal factor in producing the effect. The following experiments will show, however, that inversion must be attributed in large measure to the mineral portion as well.

Experiment 1

Here recorded are a number of soils of all types, which have significant inverting action.

To 50 cc. of a 5 per cent cane sugar solution 5 gm. of soil were added. The mixture was heated 1½ hours on a water bath at a temperature of 83° to 85°C.

[3] The action of some neutral salts in increasing the inverting power of strong acids has been of considerable interest, but will not be taken up here. See especially Spohr (89), also Acree (4) and Snethlage (86), and many others.

The quantity of invert sugar was determined in an aliquot of the filtrate by Cole's method (25), and reported in terms of grams of sucrose inverted. In another portion of the filtrate, the hydrogen-ion concentration was determined by the colorimetric method perfected by Sörensen (88, 102). The results for hydrogen-ion concentration are given in the same terms as used by Sörensen: $P(H)$, as the logarithm (base 10) of the normality with respect to hydrogen ions, the minus sign being understood. For instance, a $P(H)$ of 6.1 refers to a hydrogen-ion concentration of $10^{-6.1}$ normal. Soils are arranged in order of decreasing powers of inversion:

SOIL	SUGAR INVERTED PER 50 CC.	P (H)
	gram	
Japanese, 14	0.6179	7.07
Japanese, 11	0.1850	6.98
Japanese, 13	0.1360	7.08
Japanese, 10	0.1248	7.13
Japanese, 8	0.1197	7.14
New York, P2	0.1132	7.08
New York, 00	0.1054	7.07
New York, Turkey Hill, 0	0.0986	7.08
New York, 720	0.0980	7.08
New York, 729	0.0935	7.08
Japanese, 5	0.0890	6.98
Japanese, 7	0.0872	7.08
New York, 16207	0.0852	7.13
Japanese, 6	0.0811	6.98
New York, Dutchess Silt Loam, Orange Co., 1	0.0722	7.06
New York, P3	0.0680	7.14
New York, 1691	0.0644	7.06
New York, 1	0.0554	7.06
New York, Roberts 1	0.0545	7.14
New York, 724	0.0510	7.14
New York, Vergennes Clay, Washington County	0.0510	6.98
New York, Dutchess Silt Loam, 2	0.0482	6.98
New York, Cornell Wood Lot	0.0476	7.09
Japanese, 4	0.0476	7.13
New York, "Alfalfa"	0.0452	7.15
New York, 2109	0.0456	7.14
New York, 732	0.0442	7.14
New York, Vergennes Clay, Jefferson County	0.0421	7.14
New York, "Potato"	0.0380	7.08
New York, Turkey Hill, V	0.0300	7.14
New York, Ontario Loam, Oneida County	0.0244	7.14
New York, P1	0.0228	7.14
New York, Ontario Loam, Monroe County	0.0210	7.14
New York, 1613	0.0224	7.14
New York, 11	0.0182	7.14
New York, 6 (alkaline)	0.0000	7.35
Muck, 1713	0.0277	
Muck, 1712	0.0252	
Muck, 175	0.0210	
Muck, 1711	0.0193	

A solution of sugar made up to 5 per cent such as was used in this experiment, was found to contain 0.0026 gm. of invert sugar per 50 cc., and to have $P(H)$ of 7.07. The same solution heated for 3 hours under the conditions observed above, was then found to have 0.0036 gm. of invert sugar per 50 cc., and $P(H)$ of 7.06. It has already been mentioned that water had been found by others to have some decomposing action on sucrose; however, it is seen, that under the conditions of these experiments, this action is so small that it may be generally neglected. As a further precaution, water extracts of several soils were titrated as in Cole's procedure, to find whether or not some of the reducing power might be due to something dissolved from the soils: the mucks were found to yield slightly reducing extracts, but the mineral soils showed either none at all or very little, indeed. This factor may also be neglected, except, perhaps, where the total inversion is found to be exceedingly slight.

THE CHARACTER OF THE SOIL EXTRACTS

Bearing in mind that a solution of absolute neutrality has a hydrogen-ion concentration of $10^{-7.07}$ normal, it may be seen from the above experiment that soils can exhibit significant inverting power while their sugar extracts are only slightly acid, and in most cases neutral or alkaline. This would seem to indicate that inversion is principally a property of the solid phase—that there cannot be enough acid in solution to account for all. However, attention was given to the possibility of catalytic action being due to something in the solution phase.

In considering this possibility, there are two conditions under which it might be found to hold. First, there may be some water-soluble acid in these soils. (Hanley found frequently that the water extracts of his soils had inverting power.) Second, there may be some soil acids that are not soluble in water, yet are soluble in cane sugar solution. (There are many known cases of substances being more soluble in sugar solution than in water (100).)

If water-soluble acid is present in the soils, then water extracts should exhibit inverting action.

Experiment 2

Ten-gram portions of some of the most acid New York soils were extracted with 50 cc. of distilled water, allowing the water to stand in contact for 3 hours at 83° to 85°C. To 25 cc. of the filtrate was added 25 cc. of 10 per cent cane sugar solution, and then heated for 3 hours as usual. The amount of sugar inverted is recorded, together with the amount of sugar inverted by the same soil in mass in $1\frac{1}{2}$ hours (taken from Experiment 1). These results are corrected for the amount of invert sugar present in the cane sugar itself, and in some cases by a small figure representing the reducing power of the water extracts of the soils.

SOIL	SUGAR INVERTED BY WATER EXTRACT FROM 5 GM. OF SOIL	SUGAR INVERTED BY SOIL ITSELF (SEE EXPERIMENT 1)
	gram	*gram*
P2	0.0009	0.1132
Turkey Hill, 0	0.0026	0.0986
P3	0	0.0680
724	0.0014	0.0510
P1	0.0055	0.0228
Muck, 1713	0	0.0277
Muck, 1711	0	0.0193

It is seen that water extracts from only a few of the most acid soils have any inverting power, and that insignificant when compared with the power of the soil itself. This is in agreement with conclusions usually drawn concerning the reaction of water extracts of soils. It is probably only where air and water movement in a particular soil are restricted, or under other peculiar conditions, that water-soluble acids can form and accumulate (31). But on the other hand, water extracts of the most acid soils, when boiled to remove carbon dioxide, have been usually found to be alkaline (22, 63, 97). As will be referred to again later, the use of the hydrogen electrode in determining hydrogen-ion concentrations of soil suspensions seems, at first sight, to yield contradictory results, but it must be remembered that these measurements have been made upon suspensions and extracts in the cold and containing carbon dioxide in considerable quantities. This is an acid, and cannot but have some influence upon the hydrogen electrode, yet cannot be said to be an undesirable constituent of soil systems. At the temperatures used in digestions with cane sugar, carbon dioxide would be driven off, and could not therefore be a factor in the inversion reaction.

Consideration of the second possibility led to the following experiment.

Experiment 3

Five-gram portions of two moderately acid New York soils were heated with 50 cc. of a 10 per cent cane sugar solution at 83° to 85°C. After 2 hours the soil was filtered away, and the amount of sugar that had been inverted determined in aliquots of the filtrates: Soil A, 0.0369 gm. per 50 cc.; Soil B, 0.0563 gm. The remainder of the filtrates were further heated for 12 hours and the invert sugar again determined: A, 0.0377; B, 0.0546. Other digestions were prepared with identical conditions except that the soils remained in contact with the solutions for the entire 2 hours; the amount of sugar inverted by Soil A was 0.0561 gm., and by B, 0.0848 gm.

These results show that no continued inversion took place in the sugar extract after the soil was removed, therefore there could not have been any acid extracted. Yet when the soil was in contact with the solution the whole

time much more invert sugar was produced, indicating again that the active agency remained with the soil mass.

ACTIVITY DUE TO THE SOLID PHASE

Experiment 4. A study of the amount of inversion produced by varying quantities of the same soil

All sugar solutions were prepared containing 10 per cent of cane sugar. To 50-cc. portions, different quantities of soil were added as indicated. The time of digestions was 1½ hours, and the temperature was kept at 83° to 85°C. Results are given in grams of sugar inverted in the 50 cc.:

AMOUNT OF SOIL ADDED	SOIL P1		SOIL P3		SOIL 724	
	Invert sugar	P (H)	Invert sugar	P(H)	Invert sugar	P (H)
grams	*gram*		*gram*		*gram*	
1	0.0107	7.07	0.0340	7.07	0.0342	6.98
2	0.0159	7.07	0.0680	7.07	0.0653	6.98
4	0.0188	7.09	0.0816	7.07	0.0673	6.98
8	0.0367	7.07	0.1088	7.07	0.1061	6.98
16	0.0612	7.07	0.1061	7.07	0.1115	6.98
32	0.1295		0.1134	7.07	0.1360	6.98

According to the hypothesis of Guldberg and Waage (65), "the active masses of solid bodies which participate in a chemical equilibrium are constant." It should be specified, however, that this applies where the activity of the solid is due only to that part which passes into solution. For, if there is present a saturated solution, its concentration must, of course, be independent of the amount of solid present over and above that necessary to produce it. If the soluble part only enters into the reaction, then, barring secondary effects, the action will be the same whether the amount of solid present be much or little. But, on the other hand, if the solid phase is active in the reaction, then, the extent to which such a reaction proceeds will depend upon the amount of that solid present. If soils are considered as containing very slightly soluble acids, then the quantity of acid in solution will not be different whether 1 or 32 gm. of soil stand in contact with a given amount of water, and therefore there should be the same amount of inversion by soils whether 1 or 32 gm. are used. The above results show that this is not the state of affairs but that the amount of inversion depends upon the amount of soil present The only interpretation that can be made, then, is that the solid phase itself is the active factor in inverting cane sugar, and not the solution phase of some highly insoluble acid.

While the quantity of sugar inverted depends upon the amount of soil present in contact with the solution, it is noted that the hydrogen-ion con-

centration of the extracts is independent of this ratio—remaining the same whether 1 or 32 gm. of soil stand in contact with 50 cc. of solution. Similarly, Sharp and Hoagland (83) found "comparatively insignificant changes in the H-ion concentrations when widely varying proportions of water to soil are used." These facts bring out very strikingly that that acidity which is bound up with the soil mass is independent of the character of the solution standing in apparent equilibrium, and that such acidity cannot be measured by determining the hydrogen-ion concentration of the solution in contact with that soil.

Experiment 5

If the active agency in the inversion of cane sugar is the soil particles themselves, then the reaction must take place at the surface of the particles only. In such a case the rate of inversion would depend upon the rapidity with which the inverted sugar diffused away from this region, and with which the sugar unacted upon diffused toward it (64). It was thought that shaking might hasten the inversion, therefore, by causing the soil surfaces to come in contact with fresh solution oftener than would happen if the mixture were allowed to stand quietly. As the digestions in this experiment took place at room temperature, but little total inversion was found; however, the amount of invert sugar produced is seen to be very much greater where the samples were shaken:

SOIL NUMBER	STANDING 24 HOURS	SHAKING 9 HOURS; STANDING 16 HOURS
	gram	*gram*
1	0.0085	0.0238
2	0.0085	0.0293
6	0.0031	0.0146
9	0.0097	0.0287

Experiment 6

Here an attempt is made to lessen the inverting power of soil by repeated extractions with water and with sugar solution. Five-gram portions of soils were heated on a water bath for 2 weeks with (a) water, and (b) 5 per cent sugar solution. Every day the supernatant liquid was poured off and replaced with fresh. Finally, the inverting powers of the residues were determined, and compared with that of the same soils untreated: Soil 2 untreated, 0.2744 gm. sugar inverted; same extracted with water, 0.1945 gm.; same extracted with sugar solution, 0.1400 gm. Soil 9 untreated, 0.1689 gm.; same extracted with water, 0.1066 gm.; same extracted with sugar solution, 0.0866. Soil 12 untreated, 0.1215 gm.; same extracted with water, 0.0955 gm.; same extracted with sugar solution, 0.0982 gm. Soil 15 untreated, 0.1360 gm.; same extracted with water, 0.0867 gm.; same extracted with sugar solution 0.0870 gm.

In this as well as in other experiments, where long continued extraction of soil has been employed, it has never been found that the inverting power can

be reduced to zero, though as is seen, in some cases a very insignificant reduction can be effected. During the long continued extraction treatments at high temperatures, it is probable that many changer are taking place at the surface of the soil particles, and that the loss in inverting power is due more to this than to a dissolving away of the soil acid.

In the above experiments, many cases are found where inversion by a soil took place, and at the same time the extract was alkaline, i.e., $P(H)$ less than 7.07. This may seem strange—that there should be solution of free base, yet the soil standing in contact with this solution and presumably in equilibrium with it, should behave as an acid. The following experiment shows this even more strikingly.

Experiment 7

Five-gram portions of acid soils were treated with 0, 5, 10, 15, 20 cc., respectively, of saturated lime water, evaporated to dryness, and the inverting power of the residues tested, by digesting under the usual conditions with 50 cc. of 5 per cent sugar solution. The amount of the sugar inverted and also the hydrogen-ion concentration of the extracts are recorded:

SOIL	LIME WATER ADDED	SUGAR INVERTED	P (H)
	cc.	*gram*	
Nara	0	0.3092	6.90
	5	0.0456	7.24
	10	0.0081	7.50
	15	0.0060	7.85
	20	0.0038	8.30
Kubiki	0	0.2928	6.98
	5	0.1557	7.05
	10	0.0890	7.25
	15	0.0840	7.25
	20	0.0340	7.35
Yamaguchi	0	0.3972	6.90
	5	0.1998	6.95
	10	0.0869	6.96
	15	0.0393	7.25
	20	0.0033	8.04
Matsuyama	0	0.0668	6.98
	5	0.0150	7.35
	10	0.0075	7.50
	15	0.0069	7.65
	25	0.0018	8.30

Here it is shown again that a soil may be in equilibrium with an alkaline solution and still exhibit marked acid properties, such as inverting action on sucrose.

Thus far it has been definitely established that inversion by soil is due almost entirely to something intimately bound up with the solid particles themselves, and in very small measure indeed, to anything passing into solution.

The next experiments were prepared with the idea of testing the action on sugar, of substances, the composition or condition of which is understood, that it might be reasoned by analogy to what the inverting power of soils is due.

INVERSION BY OTHER SOLID SUBSTANCES

Experiment 8

A large number of insoluble substances were tested for their activity on cane sugar. The strength of the sugar solution was 10 per cent, 50 cc. being used, and the temperature of digestion was 83° to 85°C.:

MATERIAL	AMOUNT USED	DURATION OF DIGESTION	SUGAR INVERTED
	grams	*hours*	*gram*
Alumnia (Kahlbaum)	5.0	3	0
Silicic Anhydride (Kahlbaum)	5.0	3	0
Filter paper	0.5	2	0
Cotton	3.0	2	0
Asbestos	2.0	3	0
Sulphur	5	2	0
Fuller's earth A.	5	3	0
Fuller's earth B	5	3	0
Pumice stone	5	3	0
Charcoal	5	2	0
Bone black	5	2	0
Potter's clay	5	1½	0
Dicalcium phosphate	5	1	0.0622
Stearic Acid	5	2	0.0886
Casein	5	3	0.0425
Tannic acid	5	3	2.2356
Kaolin A	5	3	0.0358
Kaolin B	5	3	0.3060
Colloidal silicic acid*	25 cc.	3	0.0792
Humic acid†	1	3	0.4713
Barium sulfate (a)‡			0
Barium sulfate (b)‡			0.2055

* Prepared by acidifying a solution of water-glass with hydrochloric acid and carefully dialyzing until all chloride was removed.

† "Humic acid," also casein, were washed until apparently free from soluble acid.

‡ Barium sulfate (a) was precipitated in the presence of hydrochloric acid and washed until apparently free from chlorides. Barium sulfate (b), was precipitated in a neutral solution. Equal quantities of (a) and (b) were used.

Some rather unexpected results were here obtained. Fuller's earth, which has often been put forward as having characteristics similar to acid soils (63), likewise cotton (21) and charcoal (22), did not invert, neither did Kahlbaum's

silicic anhydride, though it will be shown later that in the latter case action may have been prevented by impurities. Dicalcium phosphate inverts, though the dibasic alkali phosphates do not. There was probably a decomposition of the dibasic calcium phosphate with the formation of some monocalcium phosphate which caused the inversion.[4]

If it is true that inversion is caused only by acid, then casein, tannic acid, colloidal silicic acid and humus must be themselves acids or must contain adsorbed acids. The behavior of barium sulfate would point to the latter possibility.

This led to the study of the action of materials containing adsorbed acids, and experimentation was made in many ways. It was often found that after solid materials had stood in contact with acid solutions, then washed until the washings no longer gave tests for the acid and showed hydrogen-ion concentration to be the same as that of pure water, they still had a strong inverting action on cane sugar. The following is an example of many such experiments performed, all of which will not be recorded here.

Experiment 9

Aluminum oxide was allowed to stand in contact with dilute hydrochloric acid solution; barium sulfate was precipitated in presence of an excess of hydrochloric acid, and lead sulfate in presence of sulfuric acid. All were washed with water and with cane sugar solution until the washings gave no tests for chlorides or sulfates. Action on sucrose was then tested for in the usual manner. Aluminum oxide inverted 0.0128 gm. of sugar, the extract giving a hydrogen-ion concentration of 7.24; barium sulfate, 0.0106–7.10; lead sulfate 0.0442–7.10.

Similar experiments with charcoal showed that the acid was more easily washed out, and by the time the washings became just neutral, it had no inverting action. Attempts to show that carbon dioxide adsorbed on charcoal, barium sulfate, lead sulfate and sulfur could invert sucrose gave negative results. These materials were suspended in water and carbon dioxide gas passed for a long time, but the materials thus treated would not invert. Such behavior was not anticipated, since under other conditions carbon dioxide has been found to have inverting power (2).

The above results show that solid materials can so bind soluble acids that they may not be removed with water, in any measurable degree, but still will invert cane sugar when allowed to come in contact with that solution.

ACTION OF THE ELECTRIC CURRENT IN DECOMPOSING SUBSTANCES IN SUSPENSION

If any slightly soluble salt such as tricalcium phosphate or barium sulfate is placed in the lower part of a U-tube, covered with water, and direct current

[4] In this connection see Joly and Sorel (43).

passed by immersing electrodes in the upper arms of the U-tube, it will be found that the salts are rapidly decomposed, the base moving to the cathode, and the acid in the opposite direction.

Such action of the current on silicates was observed by Brongniart and Malaguti in 1843 (20). Similar experiments on soils were performed by König, Hasenbäumer and Hassler (48), who believed that adsorbed elements, especially bases, were forced from their combination with the soil complex in this way. However, there seems never to have been made a study of the silicate residue after such treatment. In the following experiment a number of soils and allied substances were extracted with an electric current, and the behavior of the residues tested.

Experiment 10

The materials to be studied were placed in U-tubes, the arms sometimes being filled halfway. The tubes were filled with water and electrodes from a 110-volt direct current allowed to dip just under the water. From time to time the water was removed from both arms and replaced with fresh. Solutions from the cathode compartment were always found to be alkaline to phenolphthalein and that from the anode side sometimes, but not always, acid to neutral red. After the current had passed for a long time and the water had been replaced a number of times, it was found that less and less base was forced into the cathode side. Tests of the activity of the residues upon sugar were made, and compared wth the action of a similar amount of untreated material; P(H) of the sugar extracts are also recorded in the following table.

The results show that silicates can be rendered active on cane sugar by the action of the electric current, when the original substance had no such action.

In every case the solution in the cathode compartment became alkaline rapidly and strongly; acid could sometimes be detected in the anode side but was never so marked as the alkalinity of the cathode compartment. This splitting off of base together with the increase in acidity of the residue, would point to a breaking up of the silicate, silicic acid remaining behind in the residue, to which would be attributed its inverting power. On the other hand, there is the possibility that impurities might be present, such as chlorides, sulfates and phosphates, which were decomposed by the current—the bases passing into solution while the acid was adsorbed by the silicate complex. One would then attribute the inverting power of the residue to this adsorbed acid. As is seen from previous experiments, adsorbed acids do very readily invert cane sugar, and if such an adsorption of acid had taken place, this would be sure to follow. However, many investigators have shown that the silicate complex universally adsorbs base in preference to acid, so that there is the greatest reason for believing that if, for instance, tricalcium phosphate were present and decomposed by the action of the current—if the calcium were not adsorbed by the silicate, then neither would be the acid. This leaves the other alternative—that in the silicate residue insoluble silicic acid is left, after the

base with which it was in combination is driven out by the current, and it is this silicic acid which catalyzes the reaction of cane sugar inversion.

In an attempt to prepare silicic acid in such a manner as to preclude the possibility of any other acid being present, the following experiment was carried out. Starting with Kahlbaum's "silicic anhydride," which is seen to be impure, for a large amount of base may be extracted by the current, a quantity was boiled with concentrated nitric acid, filtered, and washed with water.

MATERIAL AND TREATMENT	SUGAR INVERTED PER 50 CC.	P(H)
	gram	
Soil 12, original	0.1698	7.09
Same, extracted 3 days	0.1954	7.09
Soil 10, original	0.1644	7.09
Same, extracted 3 days	0.3306	7.08
Soil 11, original	0.1884	7.09
Same, extracted 3 days	0.4094	7.09
Soil 13, original	0.1810	7.09
Same, extracted 3 days	0.2611	7.09
Orthoclase, original	0	7.27
Same, extracted 6 days	0.0320	7.14
Same, extracted 30 days	0.1040	7.09
Appophyllite, original	0	7.35
Same, extracted 6 days	0.0090	7.16
Same, extracted 30 days	0.0300	7.14
Albite, original	0	7.28
Same, extracted 6 days	0.0168	7.14
Same, extracted 30 days	0.0334	7.14
Olivine, original	0	7.24
Same, extracted 6 days	0.0050	7.16
Fluoridin, original	0	7.86
Same, extracted 3 days	0.0220	7.14
Water-glass very dilute, original	0	alkaline
Same, extracted until free from base	0.0300	7.07
Fuller's earth, original	0	7.76
Same, extracted 3 days	0.0140	7.14
Kahlbaum "silicic anhydride," original	0	7.14
Same, extracted 3 days	0.5375	7.07

A portion of this was tested by extracting with the current to find whether or not all base had been removed. The acid treatment was repeated as well as the testing until all base had been removed. The material was then dried and ignited until it showed no inverting action on cane sugar. (At this point there must have been pure silicic oxide free from base or any acid substance.) This was suspended in dilute sodium hydroxide in a beaker, the electrodes allowed to dip in the solution, and the current passed. By this treatment, it was hoped that some of the silicic oxide would be changed to sodium silicate. After several days of this treatment, the whole was transferred to a U-tube and the base removed by action of the current as already described. The

residue was then found to invert cane sugar very strongly, the extract showing a neutral reaction. That it is possible to have an insoluble silicic acid which inverts cane sugar is proven, and it must be concluded from the above considerations, that the presence of such in acid soils is a strong possibility.

Many attempts were made to prepare an "aluminic acid" in the same way, but without success. Such a substance, if it exists, does not invert cane sugar, and cannot be a factor in the inverting action of soils.

HYDROUS OXIDES ON CANE SUGAR

Lindet explained the inverting power of some metals as probably being due to the formation of hydrous oxides, which in some way liberated acid hydrogen. Copper, lead, tin, bismuth, aluminum and antimony were thus believed by him to cause inversion.

Hydrous oxides of lead, copper, bismuth, alumium, zinc and iron have been prepared, unusual precautions being observed to preclude the possibility of having impurities of adsorbed acid or base present. None has been found to have inverting power. Oxides of iron and aluminum in soils, therefore, cannot be factors in catalyzing this reaction.

INVERSION AND SELECTIVE ADSORPTION

There are many examples in colloid chemistry of solid materials selectively adsorbing or removing from solution either the base or the acid part of a neutral salt, leaving the other in solution. This is explained, by postulating that the salt reacts with water in a greater or lesser degree, depending upon the "strength" of its constituents, forming basic hydroxide and free acid. Of course, there is always the tendency for these to unite again and the reaction to go in the opposite direction. Allowing M R to represent a salt in solution, the reaction would be:

$$MR + HOH \rightleftarrows MOH + HR$$

Solid materials in contact with liquids are thought to bear electric charges at their surfaces. If the solid is negatively charged it will adsorb from salt solution the basic hydroxide, if positively charged, the acid will be removed. The supernatant liquid in the former case would be acid, and in the latter, alkaline. The removal of either base or acid would allow more salt to hydrolyze, and this action would continue until such high concentrations of acid or base had accumulated that further action would be prevented. Soils, and in fact, most solids, in contact with water bear negative charges. If they are allowed to stand in contact with neutral salts, the base is removed, and the acid accumulates in the supernatant liquid.

In systems where this absorbing effect is strong, the acid may be easily tested for by means of indicators, but where it is very slight and indicators fail, the inversion test should furnish a means for detecting the acid.

Experiment 11

To 50-cc. portions of a 10 per cent cane sugar solution, containing also 7 per cent of potassium chloride, were added various substances in amounts indicated, and digestions allowed to proceed at 83° to 85°C. for 3 hours. Similar digestions were prepared except that no potassium chloride was present. The amount of sugar inverted per 50 cc. is recorded:

SOLID PHASE	AMOUNT	SUGAR INVERTED; KCl NOT PRESENT	SUGAR INVERTED; KCl PRESENT
	grams	*gram*	*grams*
Cotton	2	0	0.0226
Sugar charcoal	5	0	0.0173
Filter paper	1	0	0.0302
Sulfur	5	0	0.0444
Asbestos	2	0	0.0943
Silicic anhydride	5	0	0.0948
Fuller's earth, no. 1	5	0	0.0475
Fuller's earth, no. 2	5	0	0
Kaolin, no. 1	5	0.0358	0.1574
Kaolin, no. 2	5	0.3060	0.9180
Casein	5	0.0425	0.2599
Humic acid	5	0.4713	0.8954
Colloidal silicic acid	25 cc.	0.0792	1.1782

These results show how it is possible to have inversion take place through the acid resulting after the selective adsorption by negatively-charged insoluble substances of the basic part of the neutral salt. In the following experiment a much lower concentration of salt is used, and the action of soils is recorded.

Experiment 12

Five grams of soil were added to 50-cc. portions of a 5 per cent cane sugar solution containing also 30 parts per million of potassium chloride. Digestions took place at 83° to 85°C. for 1½ hours. At the same time checks were run with no potassium chloride present. Results are given in grams of sugar inverted per 50 cc.:

SOIL NUMBER	SUGAR INVERTED; KCl NOT PRESENT	SUGAR INVERTED; KCl PRESENT
	gram	*gram*
1	0.2040	0.2414
8	0.1020	0.1326
9	0.0884	0.1210
10	0.1054	0.1414

In a similar manner, digestions were run in which 0.07 per cent of calcium sulfate was used instead of potassium chloride:

SOIL NUMBER	SUGAR INVERTED; $CaSO_4$ NOT PRESENT	SUGAR INVERTED; $CaSO_4$ PRESENT
	gram	*gram*
1	0.2040	0.2528
8	0.1020	0.1700
9	0.0884	0.1326
10	0.1054	0.1516

The effect of the addition of 0.1 per cent of tricalcium phosphate was tested similarly. Soil A with no addition inverted 0.0405 gm. of cane sugar, with tricalcium phosphate added, 0.0450 gm.; soil B, similarly, 0.0485 and 0.0589 gm.

Soils in contact with weak solutions of potassium chloride, calcium sulfate, or with tricalcium phosphate present, exhibit considerably greater inverting power than when alone, showing again a selective adsorption of base. Salts in such concentrations as these may always be found in soil solutions. It should be concluded, therefore, that inversion of cane sugar by soils may be due in part to the acid set free after the selective adsorption by the soil of base from any neutral salt that might be present.

It is entirely probable that a part of that acidity obtained by the hydrogen electrode in contact with soil suspensions results from such a reaction as this. In such cases, this effect might be produced by the action of some slightly soluble salts of the soil, or, which would surely be the case if considerable care were not taken in carrying out such determinations, the neutral salt (KCl) in tubes used in making contact to the calomel electrode would diffuse to the region of the soil, and an acid would then be formed, either through selective adsorption as just described, or through a modification of this phenomenon—basic exchange (77).

DISCUSSION

Significance of the inverting power of soils

In this paper it has been brought out that insoluble silicic acid and adsorbed acids, as well as soluble acids, may invert cane sugar. As it has never been proven that this reaction can be catalyzed by any other inert substance than acid, the inverting power of soils must be an indication of the actual acid there.

Probably there has never been another method used in the study of soils that measures the relative amounts of *true* acid. A possible exception is the method suggested by Conner (26), which determines the saponification of ethyl acetate in contact with soils, though he did not prove that there were not other substances in soils than acid which could cause or aid this action. The determination of hydrogen-ion concentration by the hydrogen electrode in contact with a suspension of soil in water has been used by Saidel (80), Fischer (30), Gillespie (34), Sharp and Hoagland (83), and Plummer (72).

It is claimed for such results that they represent the "intensity of acidity" rather than the "quantity of acid" present (35). At any rate, it is evident that this "intensity of acidity" as measured by the hydrogen electrode can be only that which is in the solution phase, and that this acidity is not necessarily dependent upon that acidity which is bound up with the soil solid phase. Experiments 1, 4, 7 and 10 have shown that they are not dependent. Indeed, a soil may be of such nature as to yield an alkaline solution to water, and still have acid properties. Similarly, the results obtained by Bovie (17) show that adsorbed acids are not detected by the hydrogen electrode. Therefore, acidity of soil extracts as determined by the hydrogen electrode cannot be a measure of that much larger acidity—that which does not pass into solution. On the other hand, the inverting power of soils is due chiefly to this acid—insoluble and adsorbed.

Quantitative considerations

Only in a general and comparative way may quantitative interpretations be placed on the results recorded in this paper. Quantities of cane sugar inverted by different soils under identical conditions are indicative of the relative amounts of the acids therein, but it would be impossible to draw conclusions as to the absolute amounts of acid present, since, as is seen from experiment 4, the amount of sugar inverted varies with the amount of soil in contact—and yet not in direct ratio. However, some idea may be obtained of the acid equivalent of these soils by comparing their inverting powers with that of acid of known strength under identical conditions. The following shows the amount of sugar inverted by hydrochloric acid of different strengths. Fifty cubic centimeter portions of 10 per cent sucrose solutions containing acid of strengths indicated were heated for 1½ hours at 85° to 87°C., and the amount of sugar inverted then determined: N/100 HCl, 1.815 gm. sucrose inverted; N/1000, 0.7141; N/5000, 0.2496; N/10,000, 0.1196; N/20,000, 0.0323; N/40,000, 0.0236; N/100,000; 0.0101. Comparing these figures with results obtained in experiment 1, it will be seen that 5 gm. of most of the acid soils effect greater inversion than is produced by N/20,000 acid which was found by Cameron and Breazeale (23) to be injurious to clover, and judging from the results of experiment 4 it is probable that all soils studied could be made to invert much more than this amount of cane sugar if larger quantities were used.

It is, of course, not fair to conclude that the acid present in the soil mass, and producing the inverting effect, is as injurious to plants as such a soluble and highly ionized acid as hydrochloric acid. Yet it must be recognized that while soil acid itself is not in solution, such large amounts of this acid are often present as to be potentially equivalent to a soluble acid of too high concentration to permit satisfactory plant growth. If not having a direct injury on the plant, this insoluble acid must injuriously modify the character of the soil solution.

Adsorptive power

While true insoluble acid may make for infertility, directly or indirectly, there are other properties that a soil may have that contribute to this condition, and may even be dominant. Many substances, including most soils when in contact with a solution containing a basic element, have the power of removing a part of that element from solution, and holding it more or less tenaciously against the leaching effects of water. Where the solid material contains acid, this action is due no doubt in part to it, but there are many examples recorded in the literature of colloid chemistry, where substances containing no acid exhibit this phenomenon as well.

This adsorptive force, distinct from the condition of acidity, has previously been recognized in soils. "Absorptiv ungesättigte" (74), and "negative acidity" (59) are terms aptly used to describe it. That there is not necessarily any relation between this force and acidity, is brought out especially in the behavior of muck soils. When these are tested for inverting power, relatively little is found, yet it is well known how strongly this type of soils removes free base from alkaline solution.

On account of the fact that base is found to be attracted and held through the adsorptive power of soils, and since such would be expected of an insoluble acid, it has sometimes erroneously been assumed that insoluble acid is the only cause. Some have calculated the amount of acid which would necessarily be present to unite with the same amount of base as they found soils to combine with (81, 92, 96). These figures run so high that it would be ridiculous to conclude that such amounts of acid are really there.

As to the nature of this adsorptive force, whether chemical or physical there has been much speculation (3, 8, 14, 18, 32, 94, 105 and others). However, recent views (6, 10, 38, 51) concerning valence and attraction between molecules would lead to the conclusion that there may be really no sharp dividing line between chemical union and adsorption, which is generally considered to be due to a physical attraction, all such combinations differing only in degree. With this conception, it should properly be considered that bases are held in soils in every possible degree from loose to strong combinations, the strongest corresponding with that which is found in chemical compounds, the weakest being merely adhesions to the surfaces of particles, easily broken.

Amphoteric character of soils

Very significant were the results obtained in experiments 1, 7, and 10, where soils showed inverting power, and at the same time the digestion extracts were found to be alkaline. Similarly, Hanley found some soils to have inverting power which contained calcium carbonate. There is no doubt, then, that a soil may contain an acid and at the same time enough base to neutralize this acid without this neutralization ever taking place. This phenomenon is related to the fact that acid soils have been found to neutralize acid solutions partially

when brought in contact (16, 61, 77).[5] Similarly it can be shown that the inverting power of a mixture of an acid solution in contact with an acid soil is much less than the sum of their inverting powers as obtained separately:

Digestions were prepared of N/500 hydrochloric acid and 5 per cent cane sugar, with 5 gm. of different soils, and heated for two hours at about 85°C. Amounts of sugar inverted were: Soil A, 0.9248; Soil B, 1.0562; Soil C, 0.7912 gm. The acid alone was found to be sufficient to invert 1.0084 gm. of sugar under the same conditions, while Soil A alone inverted 0.5542 gm. of sugar; Soil B, 0.3819; and Soil C, 0.3316. There was considerably less inversion by the mixtures than might have been expected from adding the inverting powers of the two. Loss for Soil A + acid was 0.6378 gm.; Soil B, 0.3341; Soil C, 0.5488.

These facts would further show that true acidity in soil and the adsorptive process are separate and distinct, adsorbed bases in acid soils being capable of neutralizing soluble acid, and unless this is recognized soil behavior must seem paradoxical. Thus we would have a material which at once neutralizes an acid, apparently neutralizes a base when in contact with a base, and also inverts cane sugar. In this respect soils might be said to be amphoteric. Hundeshagen (41) reached similar conclusions and applied this term in his work upon kaolin.

Complexity of properties of the soil mass

Owing to the complexity of the soil mass and the widely differing properties of its constituents, there may be found yet other contradictory results. One need only try out all the methods for "soil acidity" on a given set of soils to see clearly that a soil may be classified as of high acidity by one method and low by another. Basic exchange with neutral salts (27, 40), or with basic salts (44, 56); adsorption of base from a solution of base (5, 16, 97), or of dyes (48); decomposition of insoluble carbonates (93), or soluble carbonates (42, 60); liberation of iodine from a mixture of potassium iodide and potassium iodate (13, 36), or from a mixture of potassium iodide with potassium nitrate (55), or with potassium nitrite (27); saponification of ethyl acetate (26)—are phenomena which have been made use of in indicating the quantity of acidity in soils, yet it cannot be said that these are all influenced by acid alone and no other factor; many soil properties or constituents are influential here. No doubt some of these phenomena are related, and as has been pointed out by one of us (77) and Kappan (47) there may be tendencies for equilibrium to be established between the various agencies effecting them, but there is no other reason why the different methods used in determining soil acidity should be expected to give parallel results.

A given method for soil acidity cannot determine the fertility or infertility of a soil but can measure only that particular property upon which that method is based. While many recent publications have suggested this point of view (7, 31, 82), there are other cases where, if the investigator had realized this,

[5] Somewhat contradictory results were obtained by Spurway (90).

he might have more easily and perhaps more correctly explained his results. Thus, Parker (70) has recorded a number of experiments of adsorption by soils of base from solutions containing both sodium hydroxide and potassium chloride. He explained his results on the basis of a correlated action, while it is probable that base is adsorbed from each more or less independently of the other, being due to different constituents in the soil.[6] Veitch and others (63, 98) point out that the organic part of soils has properties different from the mineral part, but this cannot be the whole story—both the organic and inorganic portions of soils must be thought of as made up of a large number of distinct substances, each with its individual properties, though of course often modified by the presence of the other constituents. Along this line have been a series of researches, first by Sjollema (84) pointing to the distinctive character of different soil constituents in removing dyes from solution.

On the other hand, there is danger of inaccurate application of this idea. The distinctions between "immediate" and "continuous" lime requirement (60), also "active" and "latent" soil acidity (96), which were based upon the idea that soils were, indeed, complex and that some soil constituents reacted more quickly than others when in contact with solutions, could possibly be better explained as merely a case of some time being necessary for the complete reaction of a single constituent, or set of constituents of the soil, with the solution phase. It has been shown by a number of investigators (11, 22, 24, 71) that in ordinary reactions between solutions and solid phases in contact, while the largest part of the reaction takes place at once, much longer time is required for a complete equilibrium.

General relation of base to soil fertility

If the amount of base present in the soil is relatively small, and the adsorptive power of the soil for that base is very strong, which conditions usually go together, then the soil will be found to be infertile; those elements, especially the bases, which are necessary for plant growth are so tenaciously held by the soil mass that the plant is deprived of nourishment. Such a soil will absorb base from solutions of bases or salts, will decompose carbonates, redden litmus paper, and may still contain no acid, though it usually does contain some, as was shown in the foregoing experiments. Barring secondary effects, and in general, the addition of any mono- or di-valent base[7] will improve a soil of this nature. Most soils contain much more of every element than is necessary for growing several crops, but if the total base present is insufficient to satisfy this excessive adsorbing power, these elements are held out from the soil solution as already described. On the other hand, if any strong base is added

[6] In this connection, see also Osugi and Uetsuki (68).

[7] Mono- and di-valent bases are specified since, as will be mentioned again later, iron and aluminum act differently, even injuriously. See also Daikuhara (27).

there is a tendency for other bases to be liberated. This is the basis for the common practice of the use of soil amendments.[8]

The situation with respect to bases, however, is by no means as simple as the above discussion might imply. Aluminum and iron are bases truly enough, yet their presence in soil solution is undesirable,[9] magnesium in large amounts may be toxic (12); excess of an alkali metal may cause an unfavorable physical condition, as witnessed in soils where large amounts of nitrate of soda had been applied, and an accumulation of the corresponding carbonate resulted; it is possible that there may be particular quantitative ratios between bases that are harmful (54); there have been recorded a few instances where soils are saturated with one base and yet basically deficient with respect to another (61). Neither should it be concluded that all soil infertility is to be attributed to an unsuitable condition among the basic constituents: Toxic organic compounds are often present (87); soils may be of such poor physical character as to hinder plant growth and nourishment; as is seen in alkali soils, an excess of any soluble salt renders the soil unfit for plants; also, under peculiar conditions soluble acid in unusual and injurious amounts must be admitted (31). Yet, for all ordinary crops, the base, lime, in even excessive quantities causes no injury, and it is found that most of the above-mentioned causes of infertility are ameliorated by an application of lime.

The behavior of iron and aluminum in soils especially forms an interesting chapter. These elements may be soluble in water alone (1, 78), or become soluble through an exchange with bases from fertilizer salts (27 63, 79). While iron and aluminum are present in soil particles in larger proportions than any of the other bases, they seem not to become active unless the other bases reach a relatively low figure, and may be placed in such a condition that they cannot influence the soil solution in any way if other (stronger) bases are plentifully supplied (63, 77, 79, 82, 90). As suggested by Daikuhara, a soil is never really basically "adsorptiv ungesättig" if iron and aluminum be considered as bases. In the event that stronger bases are in low proportions, iron and aluminum may then be free to exert their influence which is injurious in many ways: (a) as already mentioned, their ions are toxic to plants, (b) if they are present in solution as salts of strong acids, these hydrolyze, producing acid, and (c) in the event that other bases are not plentiful, phosphates may revert to iron and aluminum phosphates, in which condition phosphoric acid is most insoluble, and possibility non-available to plants (91, 104) Hence, a liberal application of a base, such as lime, in addition to preventing the solubility of iron and aluminum, also furnishes a base with which the phosphoric acid may combine, and remain in a comparatively soluble condition.

[8] While this is true in general, some cases have been reported where it does not seem to old. See Lyon and Bizzell (58) and Briggs and Breazeale (19).

[9] For a review of the literature on this subject see Miyake (62).

"Acidity" is the term always applied when an infertile soil is made more fertile by the application of lime, but we must recognize that *acid* is only one of the factors causing this condition, and possibly even a minor one.

A method for the determination of relative soil acidity

Probably the only method ever developed for the determination of the true acidity of a soil depends upon the measurement of its catalytic activity upon the reaction of inversion of cane sugar. Acidity as here characterized is that which is due to soluble and insoluble acids. This is not acidity as it is ordinarily understood in soil literature and it is only one of the factors contributing toward that undesirable condition of soils found to be improved by the use of lime. Comparative measurements of true acidity of soils may be of value in some cases, when the following procedure is recommended: 5 to 10 gm. of soil is added to 50 cc. of a 5 per cent solution of cane sugar, the whole digested on a water bath at 80° to 87°C., from 1½ to 3 hours. The amount of sugar inverted is determined in the filtrate by the very delicate titrating method suggested by Cole (25).

SUMMARY

It has been shown that soils of many kinds and other insoluble materials can be made to invert sucrose; with soils this power is a property of the mineral portion as well as of the organic matter.

That this effect is due to acid cannot be doubted. This acid may be found in four different forms: (a) A slight quantity in a few soils is soluble in the sugar solution. However, the principal portion is bound to the soil particles; this may be in the nature of (b) acids which would otherwise be easily soluble, but are here strongly adsorbed on the soil particle surfaces, or (c) it may be of the nature of an insoluble acid such as silicic acid. Also, (d) a neutral salt present in the soil solution in even small quantities may be broken down while in contact with the soil mass, the basic part being more strongly adsorbed than the acid, when the latter is left free to exert its characteristic influence in inverting cane sugar.

That the inverting activity of soils is chiefly a property of the insoluble part was indicated in several ways. (a) Many soils showed inverting action on sugar in a solution which remained neutral after contact with the soil, or, in some instances, became alkaline. Also, similarly, when soil was allowed to adsorb some base, then digested with cane sugar solution, it showed inverting action, and also yielded up sufficient base to make the extract distinctly alkaline. (b) No inverting power, or very little, indeed, was found in water extracts from soils. (c) Inversion did not continue in sugar extracts after soils were removed. (d) Inversion increases with increasing amounts of soil in contact with the sugar solution, while there is no measurable change in hydrogen-ion concentration in the extract. (e) Greater inversion is produced by shaking soils with sugar solutions, than by allowing the mixtures to stand

quietly. (f) Long continued and repeated extractions of soils with water and with cane sugar solution did not greatly reduce their inverting power.

Fuller's earth, cotton, charcoal and other substances which have been described as similar to acid soils were found not to invert cane sugar.

Otherwise soluble acids so strongly adsorbed by solids as not to be removed by washing in any measurable quantity, will yet invert sugar in such condition.

Silicate minerals may be given inverting power by treating suspensions with direct current. Base is split off, passing into solution and to the cathode, and silicic acid remains with the mass, insoluble. Soil acidity is increased by a similar treatment of soils.

Contrary to previous conclusions, hydrous oxides of lead, copper, bismuth, aluminum, iron and zinc were not found to have any inverting power.

Many suspended substances having no inverting power alone, were found to produce inversion when a neutral salt was present, through selective adsorption of the base, thus setting a small amount of acid free.

"Soil acidity" is the term customarily applied when infertility of soil can be corrected by the use of a free base such as lime. There are many factors involved in causing this condition in soils, the presence of real acid being only one of them. Methods used for detecting or determining "soil acidity" generally do not measure the acid there but may depend upon many properties of soil mass in no way related to acidity. The power of a soil to catalyze the reaction of cane sugar inversion is a measure of its acid, and is probably the only method which can measure the acid bound up with the soil solid phase.

REFERENCES

(1) Abbott, J. B., Conner, S. D., and Smalley, H. R. 1913 Soil acidity, nitrification, and the toxicity of soluble salts of aluminum. Indiana Agr. Exp. Sta. Bul. 170.

(2) Abderhalden, E. 1911 Biochemisches Handlexikon, Bd. 2, p. 397. Berlin.

(3) Aberson, J. H. 1912 Das Adsorptionsvermögen der Ackererde. *In* Chem. u. Indus. Kolloide, Bd. 10, p. 13–22.

(4) Acree, S. F. 1913 On the reactions of both the ions and the nonionized forms of electrolytes. *In* Amer. Chem. Jour., v. 49, p. 345–368.

(5) Albert, R. 1909 Eine neue Methode zur Bestimmung der Bodenacidität. *In* Ztschr. Angew. Chem., Bd. 22, p. 533–537.

(6) Alexander, J. 1917 Selective adsorption and differential diffusion. *In* Jour. Amer. Chem. Soc., v. 39, p. 84–88.

(7) Ames, J. W., and Schollenberger, C. J. 1916 Liming and lime requirement of soil. Ohio Agr. Exp. Sta. Bul. 306.

(8) Armsby, H. P. 1877 On the absorption of bases by the soil. *In* Amer. Jour. Sci. Arts, s. 3, v. 14, p. 25–30.

(9) Arrhenius, S. 1889 Über die Reaktionsgeschwindigkeit bei der Inversion von Rohrzucker durch Säuren. *In* Ztschr. Phys. Chem., Bd. 4, p. 226–248.

(10) Arrhenius, S. 1912 Theories of Solutions. New Haven, Conn.

(11) Ashley, H. E. 1913 Technical control of the colloidal matter of clays. U. S. Bur. Standards, Tech. Paper 23.

(12) Aston, B. C. 1915 Magnesia and plant growth. *In* Jour. Agr. (New Zealand), v. 11, p. 493–502. *Abs. in* Chem. Abs., v. 11, p. 1811.

(13) BAUMANN, A., and GULLY, E. 1908 On free humus acids in upland moor soils and their determination. *In* Naturw. Ztschr. Land. u. Forstw. Bd. 6, p. 1–6. *Abs. in* Exp. Sta. Rec., v. 19, p. 1008–1009 (1907–08).

(14) BAUMANN, A., and GULLY, E. 1910 Die Humussäuren des Torfes. *In* Mitt. K. Bayer. Moorkulturanst., Heft 4; Ztschr. Angew. Chem. Bd. 23, p. 1760–1671. (1910). Chem. Zenthl., 5 Folge (2), Bd. 14, p. 1495–1496 (1910).

(15) BIERRY, H., HENRI, V., and RANC, A. 1911 Actions des rayons ultraviolets sur le saccharose. *In* Compt. Rend. Acad. Sci. (Paris), t. 152, p. 1629–1632.

(16) BOUYOUCOS, G. J. 1916 The freezing-point method as a new means of determining the nature of acidity and lime requirement of soils. Mich. Agr. Exp. Sta. Tech. Bul. 27.

(17) BOVIE, W. T. 1915 A direct reading potentiometer for measuring and recording both the actual and the total reaction of solutions. *In* Jour. Med. Res., v. 33, p. 295–322.

(18) BREHM, H. 1913 Über die Fortschritte und Aussichten der jüngeren Agrikulturchemie (speziell der Bodenchemie) seit Anwendung der neueren Ergebnisse der physikalischen Chemie, besonders der Kolloidchemie. *In* Koll. Ztschr., Bd. 13, p. 19–35.

(19) BRIGGS, L. J., AND BREAZEALE, J. F. 1917 Availability of potash in certain orthoclase-bearing soils as affected by lime or gypsum. *In* Jour. Agr. Res., v. 8, p. 21–28.

(20) BRONGNIART, A., AND MALAGUTI. 1843 Zweite Denkschrift über die Kaoline oder Porcellanerden, über die Natur und den Ursprung derselben. *In* Pogg. Ann. Phys. Chem., Bd. 60, p. 89–129.

(21) CAMERON, F. K. 1911 The soil solution. Chemical Publishing Co., Easton, Pa.

(22) CAMERON, F. K., AND BELL, J. M. 1905 The mineral constituents of the soil solution. U. S. Dept. Agr. Bur. Soils Bul. 30.

(23) CAMERON, F. K., AND BREAZEALE, J. F. 1904 The toxic action of acids and salts on seedlings. *In* Jour. Phys. Chem., v. 8, p. 1–13.

(24) CHRISTENSEN, H. R. 1917 Experiments in methods for determining the reaction of soils. *In* Soil Sci., v. 4, p. 115–178.

(25) COLE, S. W. 1908 Physiological chemistry, Ed. 1908, p. 44.

(26) CONNER, S. D. 1916 Acid soils and the effect of acid phosphate and other fertilizers upon them. *In* Jour. Indus. Engin. Chem., v. 8, p. 35–40.

(27) DAIKUHARA, G. 1914 Über saure Mineralböden. *In* Bul. Imp. Cent. Agr. Exp. Sta. Japan., v. 2, p. 1–40.

(28) DEERR, N. 1910 The inversion of cane sugar under the influence of acids and neutral salts. Hawaii Sugar Planters' Sta., Agr. and Chem. Bul. 35.

(29) EULER, H., AND OHLSEN, H. 1911 Sur l'inversion de la saccharose dans les rayons ultraviolets. *In* Jour. Chim. Phys., t. 9, p. 416–422.

(30) FISCHER, G. 1914 Die Säuren und Kolloide des Humus. Kühn Arch., Bd. 4, p. 1–136.

(31) FREAR, W. 1915 Sour soils and liming. Penn. Dept. Agr. Bul. 261.

(32) GANS, R. 1914 Über die chemische oder physikalische Natur der kolloidalen wasserhaltigen Tonerdesilikate. *In* Cent. Min. Geol. u. Paläontol., No. (1911) 9, p. 273–279, 299–306.

(33) GEERLIGS, H. C. P, 1898 Inwerking van neutrale Zouten op Glucose bij booge Temperatuur. *In* Arch. Jarasuikerindus., v. 6, p. 14–19. *Ales in* Deut. Zuckerindus., Bd. 23, p. 292. See Browne, Handbook of Sugar Analysis (1912 Edition), p. 667–668.

(34) GILLESPIE, L. J. 1916 The reaction of soil and measurement of hydrogen-ion concentration. *In* Jour. Wash. Acad. Sci., v. 6. p. 7–16.

(35) GILLESPIE, L. J., AND HURST, L. A. 1917 Hydrogen-ion concentration measurements of soils of two types: Caribou loam and Washburn loam. *In* Soil Sci., v. 4, p. 313–319.

(36) GREGOIRE, A. 1912 Beitrag zum Studium des Säuregehaltes des Bodens. *In* Bul. Soc. Chim. Belg., t. 26, p. 336–342, 362–375, 386–409. *Abs. in* Chem. Zent. (5 Folge), Bd. 17, (1), p. 647–648. (1913).

(37) HANLEY, J. A. 1914 The humus of acid and alkaline peats. *In* Jour. Agr. Sci., v. 6, p. 63–76.

(38) HARKINS, W. D., DAVIES, E. C., AND CLARK, G. L. 1917 The orientation of molecules in the surfaces of liquids, the energy relations of surfaces, solubility, adsorption, emulsification, molecular association, and the effect of acids and bases on the interfacial tension. *In* Jour. Amer. Chem. Soc., v. 39, p. 541–596.

(39) HARNED, H. S. 1915 The hydrogen- and hydroxlyl-ion activities of solutions of hydrochloric acid, sodium and potassium hydroxides in the presence of neutral salts. *In* Jour. Amer. Chem. Soc., v, 37, p. 2460–2482.

(40) HOPKINS, C. G., KNOX, W. H., AND PETTIT, J. H. 1903 A quantitative method for determining the acidity of soils. U. S. Dept. Agr. Bur. Chem. Bul. 73, p. 114.

(41) HUNDESHAGEN, F. 1908 Über die Anwendung organischer Farbstoffe zur diagnostischen Färbung mineralischer Substrate. *In* Ztschr. Angew. Chem., Bd. 21, p. 2405–2415, 2454–2461.

(42) HUTCHINSON, H. B., AND MAC LENNAN, K. 1914 The determination of lime requirements of the soil. *In* Chem. News, v. 110, p. 61–62.

(43) JOLY, A., AND SOREL, E. 1894 Action de l'eau sur le phosphate bicalcique. Compt. Rend. Acad. Sci. (Paris), t. 118, p. 738–741.

(44) JONES, C. H. 1913 Method of determining the lime requirement of soils. *In* Amer. Fert., v. 39, no. 11, p. 28–29.

(45) KAHLENBERG, L., DAVIS, D. J., AND FOWLER, R. E. 1899 Inversion of sugar by salts. *In* Jour. Amer. Chem. Soc., v. 31, p. 1–23.

(46) KAILAN, A. 1913 Über die chemischen Wirkungen der durchdringenden Radiumstrahlung. *In* Monatsh. Chem., Bd. 34, p. 359–364.

(47) KAPPAN, H. 1916 Causes of acidity of soils which are acid through exchange of ions. Landw. Vers. Stat., Bd. 89, p. 39–80. *Abs. in* Jour. Chem. Soc. (London), v. 110, p. 876.

(48) KÖNIG, J., HASENBÄUMER, J., AND HASSLER, C. 1911 Bestimmung der Kolloide im Ackerboden. *In* Landw. Vers. Stat., Bd. 75, p. 377–441.

(49) KULLGREN, C. 1902 Studien über die Inversion. *In* Ztschr. Phys. Chem., Bd. 41, p. 407–426.

(50) LAMBLE, A., AND LEWIS, W. C. 1915 Studies in catalysis, II. Inversion of sucrose. *In* Jour. Chem. Soc. (London), v. 107, p. 233–248.

(51) LANGMUIR, I. 1916 The constitution and fundamental properties of solids and liquids. *In* Jour. Amer. Chem. Soc., v. 38, p. 2221–2295.

(52) LEY, H. 1899 Studien über die hydrolytische Dissociation der Salzlösungen. *In* Ztschr. Phys. Chem., Bd. 30, p. 193–257.

(53) LINDET, L. 1904 Sur l'inversion du sucre. *In* Compt. Rend. Acad. Sci. (Paris), t. 138, p. 508–510.

(54) LOEW, O. 1901 The relation of lime and magnesia to plant growth. U. S. Dept. Agr. Bur. Plant Indus. Bul. 1.

(55) LOEW, O. 1909 A reaction of acid soils. *In* Ztschr. Landw. Versuchsw. Oesterr., Bd. 12, p. 461. *Abs. in* Chem. Abs. v. 3, p. 2337.

(56) LOEW, O. 1913 Studies on acid soils. Porto Rico Agr. Exp. Sta. Bul. 13.

(57) LONG, J. H. 1896 On the inversion of sugar by salts. *In* Jour. Amer. Chem. Soc., v. 18, p. 120–130.

(58) LYON, T. L., AND BIZZELL, J. A. 1916 Calcium, magnesium, potassium and sodium in the drainage water from limed and unlimed soil. *In* Jour. Amer. Soc. Agron., v. 8, p. 81–87.

(59) LYON, T. L., FIPPIN, E. O., BUCKMAN, H. O. 1916 Soils, their properties and management. New York.

(60) MACINTIRE, W. H. 1915 A method for the determination of the immediate lime requirements of soils. *In* Jour. Indus. Engin. Chem., v. 7, p. 864–867.

(61) MAC INTIRE, W. H. 1916 Factors influencing the lime and magnesia requirements of soils. Tenn. Agr. Exp. Sta. Bul. 115.

(62) MIYAKE, K. 1916 Toxic action of soluble aluminum salts upon the growth of the rice plant. *In* Jour. Biol. Chem., v. 25, p. 23–28.

(63) MORSE, F. W., AND CURRY, B. E. 1908 A study of the reactions between the manurial salts and clays, mucks and soils. N. H. Agr. Exp. Sta. 19th and 20th Ann. Rpt., p. 271–293.

(64) NERNST, W. 1904 Theorie der Reaktions-geschwindigkeit in heterogenen Systemen. *In* Ztschr. Phys. Chem., Bd. 47, p. 52–55.

(65) NERNST-TIZARD 1916 Theoretical Chemistry, p. 513, 518, 578.

(66) NEUBERG, C. 1909 Chemische Umwandlungen durch Strahlenarten II, Wirkungen des elektrischen Gleichstromes. *In* Biochem. Ztschr., Bd. 17, p. 270–292.

(67) OSTWALD, WIL. 1884 Studien zur chemischen Dynamik. III. Die Inversion des Rohrzuckers. *In* Jour. Prakt. Chem., Bd. 29 (2), 385–408.

(68) OSUGI, S., AND UETSUKI, T. 1916 Untersuchungen über die Azidität des Sauer-Mineralbodens. *In* Ver. Ohara Inst. Landw. Forsch., Bd. 1, p. 27–52.

(69) PALMAER, W. 1897 Über das Verhältniss zwischen Inversions-geschwindigkeit und Konzentration der Wasserstoffionen. *In* Ztschr. Phys. Chem., Bd. 22, p. 492–504.

(70) PARKER, E. G. 1914 Selective adsorption. *In* Jour. Indus. Engin. Chem., v. 6, p. 831–835.

(71) PINNER, L. 1915 Untersuchungen über die Ammoniak-adsorption des Bodens. *In* Kuhn Arch., Bd. 6, p. 153–238.

(72) PLUMMER, J. K. 1918 Studies in soil reaction as indicated by the hydrogen electrode. *In* Jour. Agr. Res., v. 12, p. 19–31.

(73) PLZAK, F., AND HUSEK, B. 1904 Die Inversion des Rohrsuckers, hervorgerufen durch Platinmetalle. *In* Ztschr. Phys. Chem., Bd. 47, p. 733–739.

(74) RAMANN, E. 1911 Bodenkunde, p. 242.

(75) RAOULT. *In* Jour. Fabric. Sucr. et Distill, t. 12, p. 11. See v. Lippmann, Chemie der Zuckerarten (1904 Edition), p. 1217.

(76) RAYMAN, B., AND SULC, O. 1896 Katalytische Hydratation durch Metalle. *In* Ztschr. Phys. Chem., Bd. 21, p. 481–492.

(77) RICE, F. E. 1916 Studies on soils. *In* Jour. Phys. Chem., v. 20, p. 214–227.

(78) RUPRECHT, R. W. 1915 Toxic effect of iron and aluminum salts on clover seedlings. Mass. Agr. Exp. Sta. Bul. 161, p. 125–129.

(79) RUPRECHT, R. W., AND MORSE, F. W. 1915 The effect of sulphate of ammonia on soil. Mass. Agr. Exp. Sta. Bul. 165.

(80) SAIDEL, T. 1913 Quantitative Untersuchungen über die Reaktion wasseriger Bodenauszüge. Bul. Sect. Sci. Acad. Roumaine, t. 2, p. 38–44.

(81) SCHERMBECK, VAN, A. J. 1907 Über Humussauren. *In* Jour. Präkt. Chem., Bd. 75, 517–525.

(82) SCHOLLENBERGER, C. J. 1917 Relation between indications of several lime-requirement methods and the soil's content of bases. *In* Soil Sci., v. 3, p. 279–88.

(83) SHARP, L. T., AND HOAGLAND, D. R. 1916 Acidity and adsorption in soils as measured by the hydrogen electrode. *In* Jour. Agr. Res., v. 7, p. 123–145.

(84) SJOLLEMA, B. 1905 Anwendung von Farbstoffen bei Boden-Untersuchungen. *In* Jour. Landw., Bd. 53, p. 67–69.

(85) SMITH, W. A. 1898 Über die stufenweise Dissociation zweibasischer organischer Säuren. *In* Ztschr. Phys. Chem., Bd. 25, p. 144–177.

(86) SNETHLAGE, H. C. S. 1913 Einfluss von Neutralsalzen auf katalytische Reaktionen in verschiedenen Lösungsmitteln. *In* Ztschr. Phys. Chem., Bd. 85, p. 211–262.

(87) SOILS, BUREAU OF, United States Department of Agriculture. 1907–1912. Bulletins 47, 53, 70, 74, 77, 80, 87, 88, 90.

(88) SÖRENSEN, S. P. L. 1909 Über die Messung und die Bedeutung der Wasserstoffionenkonzentration bei enzymatischen Prozessen. *In* Biochem. Ztschr., Bd. 21, p. 131–304.

(89) SPOHR, J. 1886 Über den Einfluss der Neutralsalze und der Temperatur bei der Inversion des Rohrzuckers durch Sauren. *In* Jour. Prakt. Chem., Bd. 33 (2), p. 265–284.

(90) SPURWAY, C. H. 1917 Soil acidity and the hydrolytic ratio in soils. *In* Jour. Agr. Res., v. 11, p. 659–672.

(91) STODDART, C. W. 1909 Soil acidity in its relation to lack of available phosphates. *In* Jour. Indus. Engin. Chem., v. 1, p. 69–74.

(92) STREMME, H. 1910 Über die "freien Humussäuren" des Hochmoores. *In* Ztschr. Prakt. Geol., Bd. 18, p. 389–392.

(93) TACKE, B. 1897 Über die Bestimmung der freien Humussäuren. *In* Chem. Ztg., Bd. 21, p. 174–175.

(94) TACKE, B., AND SÜCHTING, H. 1911 Über Humussäuren. *In* Landw. Jahrb., Bd. 41, p. 717–754.

(95) TREVOR, J. E. 1892 Über die Messung kleiner Dissociations-grade. *In* Ztschr. Phys. Chem., Bd. 10, p. 321–353.

(96) TRUOG, E. 1916 A new apparatus for the determination of soil carbonates and new methods for the determination of soil acidity. *In* Jour. Indus. Engin. Chem., v. 8, p. 341–345.

(97) VEITCH, F. P. 1902 The estimation of soil activity and the lime requirement of soils. *In* Jour. Amer. Chem. Soc., v. 24, p. 1120–1128.

(98) VEITCH, F. P. 1904 Comparison of methods for the estimation of soil acidity. *In* Jour. Amer. Chem. Soc., v. 26, p. 637–662.

(99) VONDRACEK, R. 1904 Beitrag zur Erklärung des Mechanismus der katalytischen Wirkungen des Platinschwarzes. *In* Ztschr. Anorg. Chem., Bd. 39, p. 24–39.
1905 Über den Einfluss der Metalle auf die Hydrolyse des Rohrzuckers. *In* Ztschr. Phys. Chem., Bd. 50, p. 560–566.

(100) VON LIPPMANN, E. O. 1904 Die Chemie der Zuckerarten. III Auf., p. 1134 and following.

(101) WALKER, J., and ASTON, E. 1895 Affinity of weak bases. *In* Jour. Chem. Soc. (London), v. 67, p. 576–586.

(102) WALPOLE, G. S. 1911 Chart presentation of recent work on indicators. *In* Biochem. Jour., v. 5, p. 207–214.
1914 Notes on regulator mixtures, recent indicators, etc. II. *In* Biochem. Jour., v. 8, p. 628–640.

(103) WHITNEY, W. R. 1896 Untersuchungen über Chromsulfat-Verbindungen. *In* Ztschr. Phys. Chem., Bd. 20, p. 40–67.

(104) WHITSON, A. R., AND STODDART, C. W. 1907 Soil acidity in its relation to lack of available phosphates. *In* Jour. Amer. Chem. Soc., v. 29, p. 757–759.

(105) WIEGNER, G. 1914 Über die chemische oder physikalische Natur der kolloiden, wasserhaltigen Tonerdesilikate. *In* Cent. Min. Geol. u. Paläontol., p. 262–272.

THE PRODUCTION OF CARBON DIOXIDE BY MOLDS INOCULATED INTO STERILE SOIL

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The purpose of the experiments reported here was to determine the physiological activity of certain molds when inoculated into sterile soil. It was hoped that such an investigation would throw some light on the question as to whether molds are active in soils and if they are, as to some of the products, chiefly carbon dioxide, of their life processes. Because of the excellent bibliography of the subject published recently by Waksman (2) we shall not go into the historical phase of the subject in detail here. In a review of the evidence and in consideration of results carried out in his laboratories, as to whether molds are present in soils in active form or not, Brown (1) shows that there is little to lead one to believe that molds are not active in soils. In this same paper, a brief review of the physiological activities of molds in soil, and their important bearing upon soil fertility, is also given.

Practically all of the work which has been done on the physiological activities of molds in soils has been carried out by using a sterile medium, usually soil, to which has been added some material, such as dried blood, in the case of a measurement of ammonification, and then inoculating with pure cultures of a mold or molds, the product sought being determined at the end of a certain period. There are two well recognized points of weakness in this procedure. First, sterilizing radically changes the soil, rendering plant-food and other material more soluble. Second, the addition of the large amounts of material (dried blood) makes the medium still further unlike typical soil conditions. The method which we used eliminated the second objection only. No method has yet been developed to render the soil sterile, and not radically alter its composition. In the last series of experiments reported in this paper, some progress was made in the solution of this problem.

EXPERIMENTAL

All the experiments were conducted with the apparatus shown in figure 1. Plate 1 gives the reproduction of a photograph of the whole arrangement of apparatus.

Experiment I

In this and the succeeding experiments, a clay loam soil with a lime requirement, by the Veitch method, of 1540 pounds of calcium carbonate per acre of

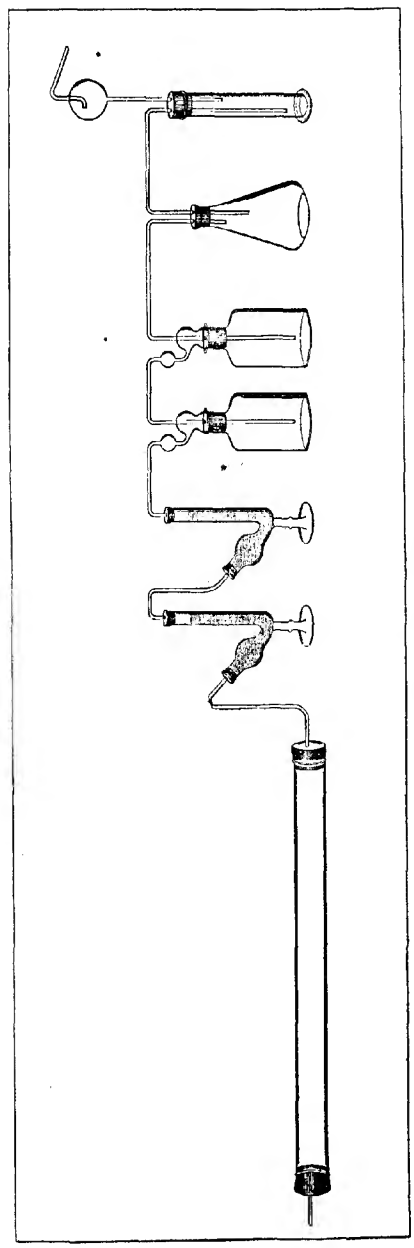


FIG. 1. DIAGRAM OF APPARATUS USED

2,000,000 pounds was used. After obtaining the soil from the field, it was partially air-dried, put through a $\frac{1}{4}$-inch sieve, and then completely air-dried and thoroughly mixed.

For Experiment I, 340-gm. ($\frac{3}{4}$-pound) portions were weighed into 24 Erlenmeyer flasks, fitted with rubber stoppers and connecting tubes as shown in figure 1. The rubber stoppers were lifted about $\frac{1}{2}$ inch out of the flasks and a large wad of cotton wrapped around the stoppers and tops of the flasks. The cotton was held in place by tying with string. The 24 flasks were then sterilized in the autoclave by steam at 10 pounds pressure for $3\frac{1}{2}$ hours, three separate times, with 48-hour periods intervening between the autoclave treatments. After sterilization was complete the 24 flasks were placed in position, the cotton wads carefully removed and the rubber stopper placed in position. The cotton plugs in the ends of the glass tubes protected the flask and contents from contamination. The flasks were then inoculated as follows. The molds were isolated from this soil by means of successive plating on beerwort agar. After pure cultures were obtained, each culture was inoculated upon a thin layer of beerwort agar in a 500-cc. Erlenmeyer flask. After the cultures thus obtained had fruited well 300 cc. of sterile water was added and then a 75-cc. portion from the suspension of each mold was transferred under sterile conditions to each of two flasks of the sterile soil. Fifteen cubic centimeters of sterile dextrose solution was added to the remaining 150 cc. of water in the flasks containing the pure mold cultures. The dextrose solution was of such a strength that the solution in the culture flasks was made exactly 1 per cent dextrose. Seventy-five cubic centimeter portions were then transferred under sterile conditions to two more flasks of sterile soil. Before lifting the rubber stoppers from the flasks to add the inoculating liquid the stopper and the top of the flasks were swabbed with an alcohol-water (1–1) solution of mercuric chloride. The proper connections were then made in the apparatus and the air current was turned on. The inoculations were made January 25, 1916, and the air was run continuously over the soil until March 9, 1916. The carbon dioxide evolved was determined by titrations at intervals shown in table 1.

After the completion of the experiment, soils from the flasks were plated out on beerwort agar to test the purity of the cultures. This was done as follows. The flask was disconnected from the apparatus with the rubber stopper and entrance and exit tubes with the cotton plugs still securely fixed. The flask was then shaken violently. This left several grams of the soil adhering to the glass tube projecting below the rubber stopper. The tube with the soil was dipped into sterile water contained in a flask, whereupon most of the soil fell from the tube. Inoculations upon the plates of beerwort agar were made in the usual way. The results are found in table 1.

In table 1 will be found the amounts of carbon given off from the various soils as carbon dioxide. The total carbon also has been computed and ap-

TABLE 1

Amount of carbon evolved as carbon dioxide from soils in Experiment. (Results expressed in pounds of carbon per acre)

NO.	TREATMENT	REMARKS	DAYS AFTER INOCULATION																												TOTAL	AVERAGE
			1	Average	2–3	Average	4	Average	5–6	Average	7	Average	8–9	Average	10–11	Average	12–13	Average	14–17	Average	18–21	Average	22–25	Average	26–32	Average	33–39	Average	40–44	Average		
1	Mucor	Pure at end	307		133		76		73		22		55		47		25		55		42		63		151		100		44		916	
2	Mucor	Pure at end	40	35	88	110	50	63	79	76	26	24	54	54	47	47	21	23	55	55	43	42	57	60	134	142	125	112	65	54	883	899
3	Mucor, Dextrose	Pure at end	276		1,770		1,480		792		224		315		329		90		247		132		117		161		108		65		6,106	
4	Mucor, Dextrose	Pure at end	139	207	960	1,365	883	1,181	1,535	1,163	371	297	378	346	376	352	136	113	374	310	194	163	145	131	167	164	103	105	65	65	5,826	5,966
5	Rhizopus	Pure at end	143		269		107		58		22		63		75		41		156		69		54		108		74		36		1,275	
6	Rhizopus	Pure at end	89	116	94	181	82	94	87	72	23	22	66	64	96	85	43	42	148	152	74	71	75	64	122	115	91	82	51	43	1,141	1,208
7	Rhizopus, Dextrose	Pure at end	163		1,300		468		100		202		477		477		118		510		321		320		127		209		78		5,770	
8	Rhizopus, Dextrose	Pure at end	159	162	631	965	472	470	630	665	128	165	306	391	256	366	115	116	199	354	238	279	171	245	225	326	205	207	62	70	3,797	4,783
9	Aspergillus niger	Contaminated	77		248		178		164		76		138		158		50		197		90		137		258		73		34		1,878	
10	Aspergillus niger	Contaminated	65	71	322	285	298	238	236	200	110	93	282	210	293	225	96	73	199	198	87	88	120	128	200	226	90	81	51	42	2,449	2,163
11	Aspergillus niger, Dextrose	Pure at end	86		626		700		675		214		658		636		136		500		129		85		102		78		46		4,671	
12	Aspergillus niger, Dextrose	Pure at end	73	79	530	578	950	825	849	762	326	270	706	682	683	659	156	146	785	642	186	157	175	132	lost	102	185	81	56	51	5,664	5,167
13	Trichoderma	Pure at end (?)	42		228		129		127		30		82		49		24		168		80		53		100		87		52		1,480	
14	Trichoderma	Pure at end	40	41	55	136	64	146	196	161	52	41	55	68	73	61	37	30	150	159	63	71	72	62	147	123	103	95	71	61	1,177	2,378
15	Trichoderma, Dextrose	Pure at end	60		578		567		1,400		306		418		343		93		475		239		163		184		115		57		4,368	
16	Trichoderma, Dextrose	Pure at end	105	82	673	625	518	542	442	921	163	234	351	384	327	335	109	101	503	489	207	223	171	167	222	103	139	127	66	61	3,996	4,182
17	Chætomium	Pure at end	19		123		65		65		28		62		75		37		184		141		107		172		151		71		1,300	
18	Chætomium	Pure at end	18	18	131	127	67	66	213	139	143	85	216	139	78	76	39	38	192	188	197	169	159	133	141	156	149	150	65	63	1,808	1,554
19	Chætomium, Dextrose	Contaminated	28		404		240		155				690		648		128		589		221		292		413		386		186		3,836	
20	Chætomium, Dextrose	Pure at end (?)	32	30	354	379	344	292	395	275	213	213	326	508	502	575	187	157	730	659	310	265	168	230	378	395	404	395	213	199	3,899	3,867
21	Soil emulsion		135		323		190		205		69		101		121		77		225		154		139		236		170		92		2,236	
22	Soil emulsion		114	124	185	254	129	159	189	197	59	64	100	100	109	115	46	61	191	208	141	147	132	135	202	219	163	166	86	89	1,849	2,042
23	Soil emulsion, Dextrose		206		960		720		1,360		378		465		316		102		492		284		287		446		297		193		6,506	
24	Soil emulsion, Dextrose		186	196	660	810	627	673	1,280	1,320	337	357	567	516	306	311	128	115	474	483	286	285	288	287	415	430	299	298	190	192	6,043	6,274

pears in the last two columns of the table. The results obtained by plating out the soils also are given in the table.

It is seen that all the culture experiments involving the Mucor *Rhizopus* were found to be pure at the completion of the experiment. The two *Aspergillus* inoculated soils numbered 9 and 10, upon plating out were found to develop nothing but *Trichoderma* colonies. Since soils 11 and 12 remained pure with respect to the colonies of *Aspergillus*, it is quite certain that the inoculating culture was pure *Aspergillus*. There is nothing to indicate at what stage of the experiment contamination took place. Sterilization might not have been complete, although that is not probable. Contamination might have taken place during the inoculation or during the passing of the air over the soils or finally during plating.

All the *Trichoderma* inoculated soils remained uncontaminated, with the possible exception of no. 13. Upon plating this soil, one plate was found to develop nothing but *Trichoderma* colonies, another plate *Trichoderma* with a single colony of a *Rhizopus*, and the third plate mainly *Trichoderma* with a single colony of *Aspergillus*. Soils 17 and 18 were found uncontaminated, soil 19 showed mainly *Rhizopus* on the plates. No. 20 upon plating, gave one plate of pure *Chætomiun* colonies, another mainly *Chætomium* and a single *Rhizopus* colony, and the third mainly *Chætomium* with a single *Aspergillus* colony. Here, as with soil 13, it seems probable that the contamination was brought about during the plating.

The results obtained on the evolution of carbon, as carbon dioxide, will be discussed now. To facilitate comparison, the average amounts of carbon given per day for each period have been computed, and the results so obtained have been plotted, and appear in figure 2. Only the results for the soils with no dextrose have been plotted.

Perhaps the most noteworthy thing to be observed is the very rapid increase in the amount of carbon dioxide given off during the second and third day, and the fourth day. In every case, except the *Chætomium* cultures, there was a decrease on the fifth and sixth days. From the manner of inoculation, very great numbers of spores were added to the soils, and the rapid increase in amount of carbon dioxide evolved is no doubt due to the development of these spores. It has been found (3) that spore formation of fungi is accompanied or closely followed by a development of large amounts of ammonia. Probably there is also a development of a correspondingly large amount of carbon dioxide and our data might be taken to prove that, but there are other factors which enter in, which may have more influence. Sterilization of soil renders organic matter, and other nutrient material, more soluble, and therefore on the addition of large numbers of fungi spores, they immediately find a suitable medium for growth, hence they all develop vigorously. Later this nutrient medium becomes partially exhausted. It is seen from figure 2 that on the eleventh day, two of the fungi, *Aspergillus* and *Rhizopus*, give an increased amount of carbon

over the previous period, while all of them give increased amounts during the period from the fourteenth to the seventeenth. This experiment which was carried out previous to the publication in regard to sporulation and ammonia

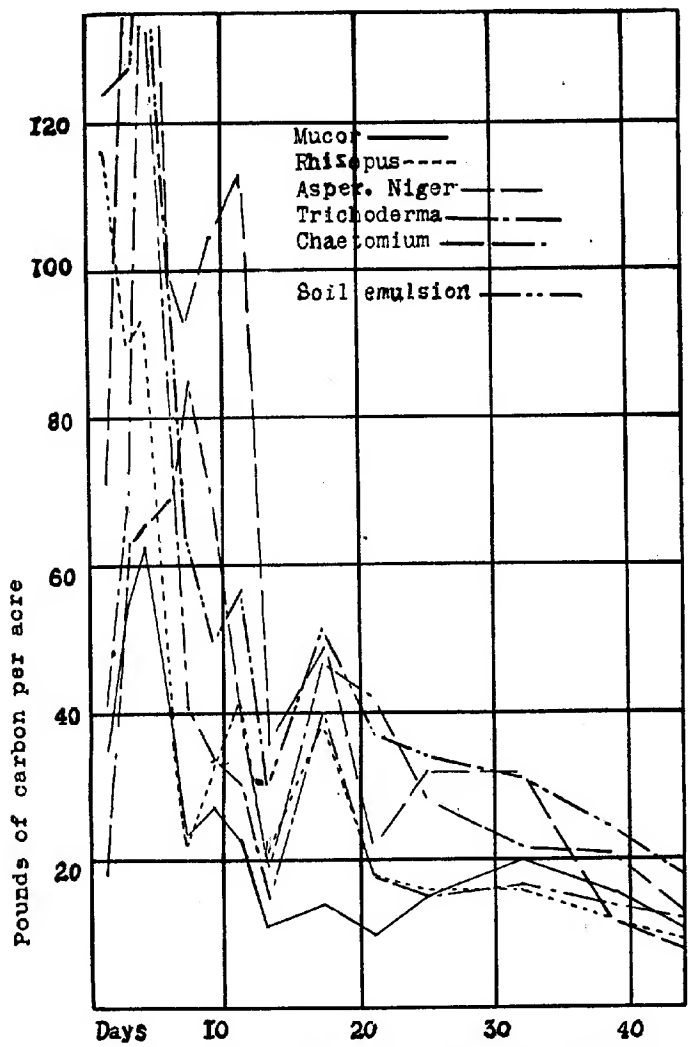


FIG. 2. DIAGRAM SHOWING AMOUNTS OF CARBON PRODUCED IN EXPERIMENT I.

production (4), was subjected to the changing temperature of the laboratory, and hence many of the fluctuations observed were no doubt due to this factor. In general, the most valuable conclusion to be drawn from a consideration of

these results, as they appear in the table and in the curve, is that in some cases the fungi gave off more carbon than the soils inoculated with soil emulsion. In other cases, less is given, but in general the order of magnitude of the amounts is the same. More in regard to the significance of this will be said later.

The results for the soils to which dextrose was added, are in general similar to the results for the soil alone, except for the larger amounts of carbon given off. When the experiment was planned, it was not known whether or not the soil alone would support sufficient mold growth to give appreciable amounts of carbon dioxide, and this was the reason for using the dextrose.

At the end of the experiment, the soils were immediately air-dried, and later were analyzed for nitrate, ammonia, and soluble non-protein nitrogen.

Since the publication of the investigation which resulted in the proposal of the method for soluble non-protein nitrogen of soil (2), we have found that practically all of the nitrogen which is extracted by the 1 per cent acid belongs to this class of compounds. For example, in an examination of several soils, from 96 to 100 per cent of the nitrogen in the acid extract was not precipitated by 2.5 per cent trichloracetic acid. Therefore, the nitrogen extracted by the 1 per cent acid is always included in the soluble non-protein nitrogen group of compounds.

It should be stated that all results published by us on the soluble non-protein nitrogen except those in the original communication, have included in them the nitrogen of the acid extract. The value of the results in the original paper are in no way impaired by their incompleteness in this respect. A paper amplifying our first communication on soluble non-protein nitrogen is in course of preparation.

The results of these determinations are given in table 2.

The sterilized soil contained 0.895 per cent of the total nitrogen as nitrate nitrogen. The mold-inoculated soils in every case contained less nitrate nitrogen, and the dextrose-treated soils usually less than the corresponding untreated soil. These facts are in harmony with the findings of others, namely, that molds may reduce or utilize nitrates, and that so far no molds have been found which bring about nitrification. It is seen that there was a considerable accumulation of nitrates in the soil-emulsion-inoculated soils, except of course in the dextrose-treated flasks.

In the sterilized soil, 3.01 per cent of the soil nitrogen was present as ammonia. In the soils treated with the *Rhizopus* and the *Chætomium*, this amount was increased, while with the other molds, and the soil emulsion the ammonia was decreased. This decrease should not be interpreted as meaning that fungi might actually use more ammonia in the field than they would cause to be formed. The conditions would be quite different if nitrifying organisms were present to utilize the ammonia as it was produced. Higher plants would in some cases also function similarly. The reason for the great reduction in the amount of ammonia in the dextrose soils, is no doubt due to the greatly en-

hanced growth of the molds, which causes them to utilize a larger proportion of more easily available nitrogen present. Another factor should be taken into consideration in regard to the dextrose-treated soils. Undoubtedly there was a great initial increase in acidity from its decomposition and this would tend to make the growth and metabolism of the molds quite different from what would otherwise be the case.

TABLE 2

Nitrate, ammonia, and soluble non-protein nitrogen in the sterilized and unsterilized soil and in the sterilized inoculated soil after the completion of the experiment

NUMBER	TREATMENT	PER CENT OF TOTAL NITROGEN		
		Nitrate	Ammonia	Soluble non-protein
Original soil		0.325	0.60	14.45
Original soil	Sterilized	0.895	3.01	22.8
1, 2	Mucor	0.67	2.77	23.8
3, 4	Dextrose, Mucor	0.68	1.57	22.2
5, 6	Rhizopus	0.59	3.08	22.9
7, 8	Dextrose, Rhizopus	0.29	0.70	18.0
9, 10	Aspergillus	0.44	1.54	21.6
11, 12	Dextrose, Aspergillus	0.39	1.30	20.4
13, 14	Trichoderma	0.54	2.58	21.0
15, 16	Dextrose, Trichoderma	0.32	0.45	19.2
17, 18	Chætomium	0.65	3.64	21.9
19, 20	Dextrose, Chætomium	0.41	1.26	19.6
21, 22	Soil emulsion	2.98	2.56	18.9
23, 24	Dextrose soil emulsion	0.74	2.25	19.3

The sterilized soil contained 22.8 per cent of its nitrogen as soluble non-protein nitrogen. There was a slight increase in the amount caused by the *Mucor* and *Rhizopus*, while all the other fungi caused decreases. The greater the amount of the soluble non-protein nitrogen present, the less, of course, the amount of complex protein compounds, and therefore of soluble non-protein nitrogen present in this experiment will give an indication as to the amount of protein material synthesized or broken down. It is probably significant that those molds which give off the greater amount of carbon have apparently caused the synthesis of greater amounts of protein material. It is also seen that the soils treated with dextrose have in every case synthesized more protein material than their corresponding untreated soil. The reverse is true in the soil-emulsion-inoculated soils. In these soils bacterial action plays, no doubt, a large part. The synthesis of more protein complexes in the soil-emulsion-inoculated soils is to be explained by the fact that there are a multiplicity of varieties of organisms active in these soils.

Experiment II

In this experiment exactly the same arrangement of apparatus was used as in the previous experiment. The same fungi were used. Instead of employ-

ing dextrose, however, lime was used. This soil, as stated above, had a lime requirement by the Veitch method of 1540 pounds of calcium carbonate per acre. In order to make sure that sufficient lime should be added to the soils to render them basic, $\frac{3}{4}$ pound of the dry soil was mixed, with sufficient lime to give in one case an excess of $\frac{1}{2}$ ton and in the other of 1.6 tons, per acre. The soils were then heated in the autoclave 3 hours per day on three alternate days, and they were then tested with litmus paper. Since there was some question as to whether the soils receiving $\frac{1}{2}$ ton of lime in excess were alkaline, it was decided to use 1.6 tons of excess lime.

Twenty-four flasks were prepared as in Experiment I. To 12 flasks, lime in the amount indicated above was added, and the 24 flasks were sterilized simultaneously by heating 3 hours in the autoclave at 10 pounds pressure on three alternate days. After sterilization of the soils, the flasks were connected with the remainder of the apparatus, and 50 cc. of sterile water was added to each flask. Inoculation was not made until 15 days after this, air being forced over the soil, and into the alkali towers. This procedure was followed in order to make sure that sterilization was complete. The amount of carbon evolved in this preliminary period is given in table 3. On the fifteenth day, inoculation was made as before, except that only 20 cc. of the inoculum was used. This gave a final moisture content of the soil about the same as that of the previous experiment. Through an oversight, the soils were not plated out at the completion of the experiment; therefore, we cannot be certain that the cultures remained pure through the course of the experiment. It is thought that pure cultures remained in all the soils, for the following reasons. Table 3 shows that from the eleventh to the fourteenth day, of the 14-day period previous to inoculating the soils, no carbon was evolved from the unlimed soils. The amounts given during the first twelve days must have come from carbon dioxide held in the soil atmosphere and soil solution. The carbon dioxide, no doubt, came from the action of the heat on the soil organic matter. If any organisms had been present they soon would have multiplied enormously with a consequent rise in carbon-dioxide production. It is seen that some carbon dioxide was still being evolved from the limed soils during the last four days of the preliminary period, but only a small amount when compared with that given during the first 12-day period. Therefore, in the case of the limed soils, it seems safe to conclude that sterilization was complete. Successive platings of the inoculating cultures of molds invariably showed them to be pure. In all cases in this experiment, and the previous one on those soils inoculated with molds, the growth appeared on the surface of the soil a few days after inoculation. In the previous experiment on the soils where the molds remained pure throughout the experiment, no evidence of a foreign growth was ever seen, while in the few cases where the plating showed the presence of invading organisms, this was evident by a superficial examination of the soil during the course of the experiment. In this second experiment, the soil always showed evidence of the mold with which it had been inoculated, and in

TABLE 3

Amount of carbon evolved as carbon dioxide from soils in Experiment II. (Results expressed as pounds per acre)

NO.	TREATMENT	DAYS AFTER PLATING			DAYS AFTER INOCULATION																														TOTAL	AVERAGE
		1-9	11-12	13-14	1	Average	2-3	Average	4	Average	5	Average	6-7	Average	8-9	Average	10-12	Average	13-14	Average	15-18	Average	19-25	Average	26-32	Average	33-39	Average	40-47	Average	48-54	Average	55-61	Average		
1	Mucor	37	0	0	4		238		134		79		93		83		122		62		49		86		63		52		64		35		39		1,203	
2	Mucor	37	0	0	12	8	210	224	76	105	68	73	79	86	77	80	124	123	78	70	65	57	122	104	92	77	74	63	80	72	44	39	52	45	1,253	1,228
3	Mucor, Lime	170	0	0	33		175		67		65		87		139		220		90		85		58		76		71		76		42		60		1,344	
4	Mucor, Lime	284	1	0	10	21	147	161	75	71	67	66	87	87	150	134	143	183	54	72	42	63	899	73	76	76	59	65	63	69	35	38	58	59	1,155	1,247
5	Rhizopus nigricans	53	2	0	22		207		76		49		57		52		64		35		35		98		40	25	25		34		32		24		848	
6	Rhizopus nigricans	36	0	0	14	18	254	230	73	74	45	47	72	64	61	56	67	65	36	35	26	30	51	74	37	38	25	25	34	34	20	26	23	23	838	843
7	Rhizopus nigricans, Lime	362	14	10	12		189		72		57		71		195		122		44		35		208		92		28		95		50		49		1,319	
8	Rhizopus nigricans, Lime	169	7	0	15	13	204	196	67	69	52	54	72	71	65	130	80	101	35	49	26	30	50	129	40	66	27	27	41	68	28	39	30	39	832	1,075
9	Aspergillus niger	50	1	0	31		446		167		134		197		125		91		35		27		77		38		35		49		40		39		1,531	
10	Aspergillus niger	27	0	0	16	23	478	462	179	173	171	152	192	194	113	119	78	84	34	34	20	23	34	56	30	34	25	30	35	42	19	28	55	47	1,479	1,505
11	Aspergillus niger, Lime	371	28	10	92		625		106		77		77		74		81		42		41		121		51		43		44				32		1,538	
12	Aspergillus niger, Lime	415	28	10	33	62	491	558	121	113	103	90	95	86	73	73	76	78	33	37	25	33	64	92	38	44	34	38	44	44	32	28	47	39	1,301	1,419
13	Trichoderma	23	0	0	11		529		278		229		279		141		99		49		43		103		47		41		54		24		42		1,976	
14	Trichoderma	27	0	0	8	9	464	491	304	291	262	245	243	261	107	124	103	101	58	53	56	49	102	102	55	51	44	42	51	52	17	24	40	41	1,914	1,945
15	Trichoderma, Lime	458	27	14	105		504		149		138		197		149		130		61		52		120		60		51		61		46		49		1,872	
16	Trichoderma, Lime	347	0	0	10	57	330	417	271	210	244	191	215	206	106	127	97	113	66	63	57	54	118	119	65	62	55	53	64	62	34	40	46	47	1,878	1,925
17	Chætomium	26	0	0	2		79		67		65		89		100		102		72		66		125		75		54		58		34		33		1,018	
18	Chætomium	27	0	0	2	2	31	55	52	59	65	65	107	98	91	95	110	106	65	68	58	62	176	150	83	79	61	57	60	59	31	32	35	34	1,030	1,024
19	Chætomium, Lime	339	0	0	6		90		54		52		77		74		86		56		46		84		59		44		47		34		28		827	
20	Chætomium, Lime	313	2	0	17	11	100	95	58	56	57	54	101	89	87	80	181	133	76	66	5	49	132	108	75	67	61	52	79	63	24	38	74	51	1,202	1,014
21	Soil emulsion	26	0	0	4		26		167		160		334		248		225		83		108		235		136		121		150		52	87	139		2,225	
22	Soil emulsion	24	0	0	2	3	140	83	160	163	202	181	315	324	205	226	180	202	88		82	95	186	210	136	136	122	121	155	152	86	87	142	140	2,201	2,213
23	Soil emulsion, Lime	395	62	31	50		341		121		136		190		134		168		115	85	99		19		141		136		165		94		134		2,221	
24	Soil emulsion, Lime	383	43	17	17	33	285	313	191	156	158	147	160	175	155	144	165	166	112	113	112	105	238	217	159	150	139	137	168	166	97	95	126	130	2,282	2,251

no case was a foreign mold growth apparent. Of course, this is not absolute proof in regard to molds and no proof at all in a consideration of bacteria. However, it would seem probable that if foreign molds were kept out, bacteria would be also.

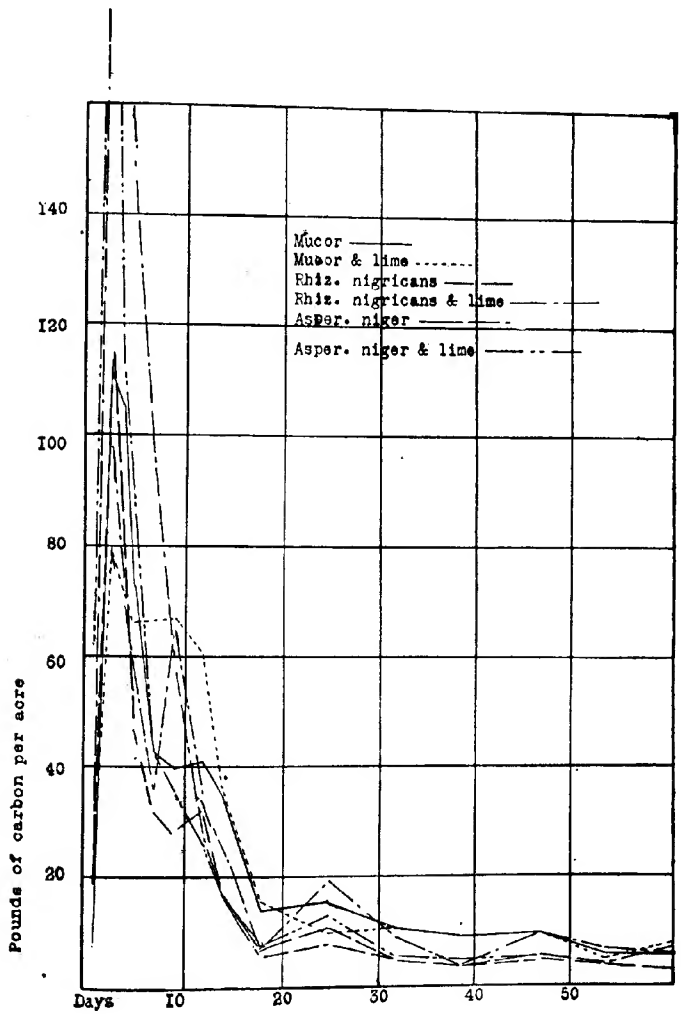


FIG. 3. DIAGRAM SHOWING AMOUNTS OF CARBON PRODUCED IN EXPERIMENT II

The amounts of carbon dioxide given from the various soils are set forth in table 3. The average amount evolved per day for each period has been computed, and the results so obtained were used in plotting the graphs in figures 3 and 4.

From the figures given in the last two columns of table 3 it is seen that the duplicates did not check very well in some cases, particularly for soils 7 and 8, and 19 and 20. No explanation of these discrepancies is apparent although

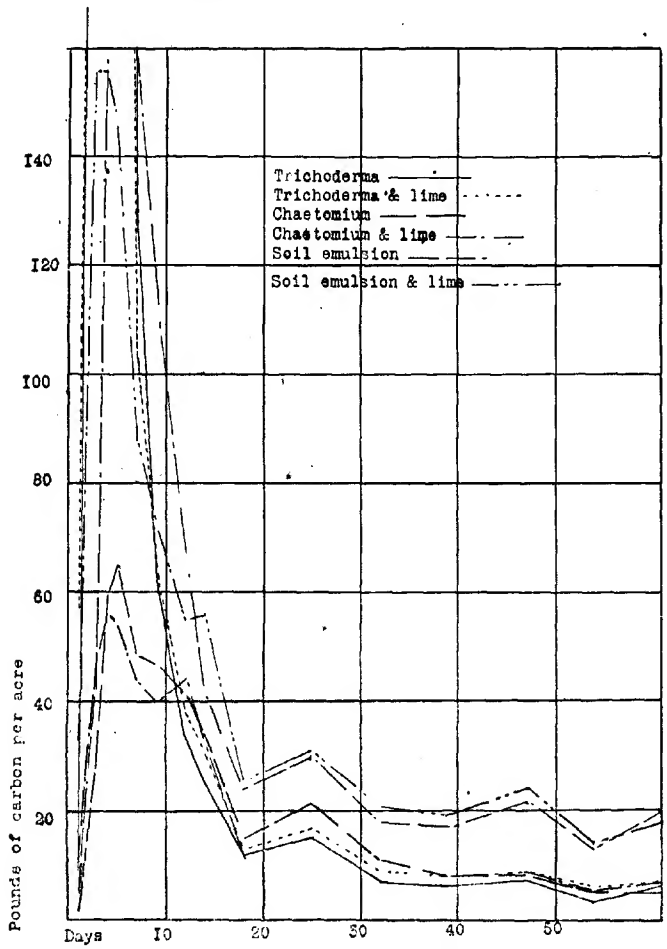


FIG. 4. DIAGRAM SHOWING OTHER AMOUNTS OF CARBON PRODUCED IN EXPERIMENT II

it is to be noted that almost invariably the unlimed soils have given more closely-agreeing duplicates than the limed. This might have been due to a difference in the amount of the lime decomposed by the heat treatment. Unfortunately, the soils were not analyzed for residual lime after the sterilization.

Keeping in mind the variations noted, it is seen that not much difference is to be noted in the amounts of carbon evolved from the limed and the unlimed soils. It is generally conceded that molds are more active in acid media than in neutral or alkaline media, yet this experiment does not seem to confirm it. The soil used had only a rather low lime requirement, so that possibly the hydrogen-ion concentration was not much different in the acid and in the limed soils. It is also seen that there is not much difference in the amount of carbon given from the limed and unlimed infusion-inoculated soils. In figures 3 and 4 the average daily results for the evolved carbon have been plotted. The course of the mold activity can be followed very easily by a study of these curves.

TABLE 4

Nitrate, ammonia and soluble non-protein nitrogen in the sterilized and unsterilized soil and in the sterilized inoculated soil after the completion of the experiment

NUMBER	TREATMENT	PER CENT OF TOTAL NITROGEN		
		Nitrate	Ammonia	Soluble non-protein
Original		0.325	0.60	14.45
Original	Sterilized	0.895	3.01	22.8
1, 2	Mucor	0.53	4.74	21.8
3, 4	Lime, Mucor	0.53	4.65	21.8
5, 6	Rhizopus	0.51	4.08	22.8
7, 8	Lime, Rhizopus	0.54	3.90	21.6
9, 10	Aspergillus	0.24	3.22	21.7
11, 12	Lime, Aspergillus	0.28	2.92	20.6
13, 14	Trichoderma	0.26	1.90	19.9
15, 16	Lime, Trichoderma	0.26	3.18	20.2
17, 18	Chætomium	0.46	4.64	21.9
19, 20	Lime, Chætomium	0.51	4.02	20.4
21, 22	Soil emulsion	0.61	5.30	19.6
23, 24	Lime, soil emulsion	4.52	2.08	14.9

The results for the determination of nitrate, ammonia, and non-protein nitrogen are given in table 4. Considering the results for nitrates, first, it is seen that in the mold-inoculated soils in every case, there is a diminution in nitrates, with the lime having no apparent effect. The soil-emulsion-inoculated and limed soils gave a considerable increase in nitrate nitrogen. There were some rather marked changes in the ammonia content of the soils. In only one case, that of the soil inoculated with *Trichoderma*, is there any considerable difference between the limed and unlimed soils, the latter having less ammonia than the limed soil. In the case of the soil-infusion-inoculated soils, no doubt the smaller amount of ammonia in the limed than in the unlimed soils is due to the greater nitrification in the former soils. The soluble non-protein nitrogen does not show any consistent changes except in case of the soil-emulsion-inoculated soils.

Experiment III

A different plan of sterilizing the soil was used in this experiment than in the preceding. It was thought that possibly sterilizing the soil with flowing steam in the Arnold sterilizer would be as effective as heating in the autoclave, and that less change would be brought about in the soil. In order to determine the number of times it was necessary to heat the soils in the Arnold sterilizer to bring about complete sterilization, the following preliminary experiment was carried out.

Several 500-cc. Florence flasks were prepared, each containing 1 pound of the air-dry soil, to which had been added 25 per cent of water, and then stoppered with cotton. It was found by experiment that it took about 40 minutes' heating in the sterilizer to bring the temperature in the center of the soil in the flask to that of boiling, namely, about 99°. Therefore, several of the flasks prepared as above were placed in the Arnold sterilizer, and heated for 1 hour and 40 minutes. This was repeated every alternate day until six heat treatments had been applied. Two of the flasks were then removed and allowed to stand at room temperature for one week, when the flasks were shaken, a few grams of the soil transferred to sterile water, and plates poured. Flasks receiving 7, 8 and 9 heat applications were treated in the same way with the following results:

6 treatments	5 contaminated plates, 3 sterile
7 treatments	1 contaminated plate, 7 sterile
8 treatments	No contaminated plate, 8 sterile
9 treatments	No contaminated plate, 9 sterile

It is seen that the soils heated 8 or 9 times in the sterilizer are undoubtedly sterile. Therefore, it was decided to heat the soils 9 times in the Arnold sterilizer. Twelve flasks were prepared, each containing 1 pound of the air-dry soil, and to six of these sufficient lime was added to give a treatment of 4 tons per acre. Twenty-five per cent of water was added, and rubber stoppers inserted bearing delivery tubes exactly as in the two preceding experiments. The flasks were then sterilized as indicated above.

Twelve of the flasks, of which six were limed, were then set up in much the same way as in the preceding experiment, except that the flasks were all placed in a water bath. It was originally planned to regulate the temperature, but after a few days, the mechanism failed. The water, however, served to keep the temperature of all the flasks alike. The inoculations were made as previously. The treatments, plating and carbon results are given in table 5.

It is seen that those flasks which had been inoculated with the *Rhizopus* were all contaminated with a *Penicillium* organism. The two unlimed soils were only slightly contaminated, while the two limed apparently were infected predominately with *Penicillium*. Although the inoculating cultures gave no indication of being impure, yet the conclusion seems rather clear that the

TABLE 5

Amount of carbon evolved as carbon dioxide from soils in Experiment III

(Results expressed as pounds per acre)

NO.	TREATMENT	REMARKS	DAYS AFTER STARTING			DAYS AFTER INOCULATION														TOTAL	AVERAGE
			1st	2–7	8	1	2	3	4–5	6–7	8–9	10–12	13–16	17–20	21–26	27–33	34–40	41–46	47–53		
1	Rhizopus nigricans	1–4	5	12	0	3	43	55	214	86	53	125	105	92	96	89	78	86	87	1,212	
2	Rhizopus nigricans	Contaminated with a Penicillium	5	8	0	3	49	72	236	78	53	113	107	88	90	85	66	82	84	1,206	1,209
3	Rhizopus nigricans, Lime		34	133	32	47	53	60	125	99	57	lost		7							
4	Rhizopus nigricans, Lime		31	100	27	48	52	55	115	77	50	106	97	78	53	68	43	55	66	963	963
5	Aspergillus niger	Pure at end	5	6	0	81	93	105	166	89	44	85	65	50	60	49	37	68	39	1,031	
6	Aspergillus niger	Pure at end	5	14	0	77	91	95	149	82	45	74	79	82	74	53	39	81	39	1,060	1,045
7	Aspergillus niger, Lime	Pure at end	34	120	32	127	91	66	83	66	39	78	82	75	60	63	56	114	118	1,118	1,118
9	Soil emulsion		4	10	0	3	27	79	322	220	113	161	224	162	97	79	67	83	58	1,695	
10	Soil emulsion		5	10	0	3	8	23	266	230	142	193	180	134	118	90	75	126	124	1,712	7,103
11	Soil emulsion, Lime		40	230	46	70	85	81	156	111	74	181	151	133	134	106	78	88	84	1,532	1,532

noculum was the cause of the contamination. Soil 3 became contaminated on the ninth day, as a result of the stopper of the flasks coming out. Flasks 8 and 12 were broken during the course of the experiment.

The results for the carbon-dioxide determinations are not markedly different from those of the preceding experiment. Because of contamination, no conclusion can be drawn from soils 1 to 4. The limed soils inoculated with *Aspergillus niger* gave off more carbon dioxide than the unlimed, but unquestionably part of the increase was due to carbon dioxide in the soil water freed from the lime. This is indicated by the fact that considerable carbon dioxide was given from the limed soils before inoculation. Also, the excess carbon dioxide given from the limed soils at the end of the first day after inoculation must not

TABLE 6

Nitrate, ammonia and soluble non-protein nitrogen in the sterilized and unsterilized soils and in the sterilized inoculated soils after the completion of the experiment

NUMBER	TREATMENT	PER CENT OF TOTAL NITROGEN		
		Nitrate	Ammonia	Soluble non-protein
Original		0.325	0.60	14.45
Original	Sterilized	0.24	2.15	21.9
Original	Lime, sterilized	0.27	3.18	19.9
1, 2	Lime, sterilized	0.22	2.51	
4	Lime, sterilized	0.27	3.64	
5, 6	Lime, sterilized	0.21	2.17	
7	Lime, sterilized	0.22	4.04	
9, 10	Lime, sterilized	0.25	3.18	
11	Lime, sterilized	1.36	2.51	

have been due to increased organic activity, but rather to the carbon dioxide in the soil atmosphere and moisture. The limed soils, both the uninoculated and the inoculated, were analyzed for residual carbonate, but practically none was found. Therefore, the heat treatment caused all the calcium carbonate to be decomposed, and no doubt considerable excess carbon dioxide was left in the soil water and atmosphere.

In table 6 the nitrate, ammonia, and non-protein nitrogen results for the sterilized and unsterilized soils are given, together with the nitrate and ammonia results for the inoculated soils.

Comparing the results on the sterilized uninoculated soils given in table 6 with the corresponding results for the same soil in table 4, it is seen that the sterilizing in the Arnold sterilizer had but little less effect on the nitrogenous compounds than did autoclave treatment. The nitrate was considerably less changed, the ammonia not much different, nor was the soluble non-protein nitrogen changed to a much less extent. Evidently, the long continued treatment at the temperature of boiling has about the same effect as the shorter but higher temperature treatment in the autoclave. It is thought that possibly

complete sterilization could be effected by a considerably less number of treatments in the Arnold sterilizer than the nine to which we subjected our soils.

The results for the nitrate and ammonia nitrogen in the inoculated soils are about the same in a general way as those for the preceding experiment, and will not be discussed further.

SUMMARY AND CONCLUSIONS

The results of the experiments reported in this paper do not give a definite answer to the question as to whether molds are active in normal soils or not. Nevertheless, the fact that in some cases more, and in all cases nearly as much, carbon dioxide is evolved from sterilized soils inoculated with molds and from sterilized soils inoculated with soil emulsion, seems indicative that they may be. Of course, in sterilized soils conditions are quite different from those in normal soils. More plant-food and other materials are in a soluble state, a condition which is known to favor mold growth. However, it hardly seems probable that sterilization would change the soil from a medium absolutely unsuited to mold growth to a medium supporting molds to an extent equal or nearly equal to the support of the whole soil flora.

The definite conclusions to be drawn from this investigation are as follows:

1. Typical soil molds when inoculated into sterilized soil grow with a vigor equal to or nearly equal to the growth induced by an inoculation with the entire soil flora, the evolution of carbon dioxide being the measure of the vigor of growth.

2. Calcium carbonate in this soil causes no marked increase or decrease in the growth of molds as measured by the evolution of carbon dioxide.

3. Sterilization in the autoclave increased the nitrate, ammonia, and soluble non-protein nitrogen.

4. Sterilization in the Arnold sterilizer increased the ammonia and soluble non-protein nitrogen to a less extent than did sterilization in the autoclave. The amount of nitrate was decreased by treatment in the Arnold sterilizer.

5. Molds in all cases caused a diminution in the amount of nitrates. Ammonia was not much changed in amount. In nearly every case there was a decrease in the amounts of soluble non-protein nitrogen.

REFERENCES

(1) Brown, P. E. 1917 The importance of mold action in soils. *In* Science, n.s., v. 46, p. 171–175.

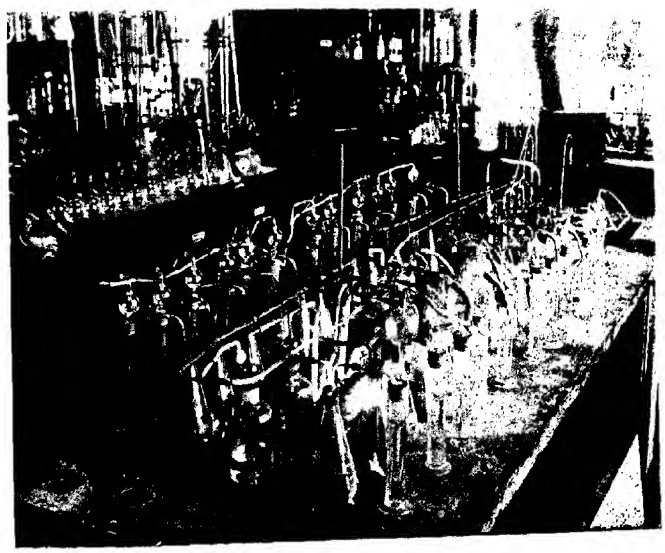
(2) Potter, R. S., and Snyder, R. S. 1916 Soluble nonprotein nitrogen of soil. *In* Jour. Agr. Res., v. 6, p. 61–64.

(3) Waksman, S. A. 1916 Soil fungi and their activities. *In* Soil Sci., v. 2, p. 103–156.

(4) Waksman, S. A., and Cook, R. C. 1916 Incubation studies with soil fungi. *In* Soil Sci., v. 1, p. 275–284.

PLATE I

APPARATUS USED IN CONDUCTING EXPERIMENTS HERE REPORTED



THE CHEMICAL EFFECTS OF CaO AND $CaCO_3$ ON THE SOIL

Part I. The Effect on Soil Reaction

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The question of the chemical and physical effects on the soil of the different forms of lime has always been a subject of great interest to agricultural investigators. Innumerable field experiments have been carried out in order to determine how crops respond to the application of different forms and amounts of lime. In the majority of cases, however, no attempt has been made to discover the precise chemical changes induced in the soil system through the addition of lime. During the course of an extensive investigation on water extracts (1, 7), osmotic pressures (2), and reactions of soils (6), the suggestion was made that it would be of considerable interest to obtain further data concerning the effects of lime on the soil as evidenced by these methods of study. The present paper deals with the reaction of the soil, while the following article takes up the questions of water extracts and osmotic pressures.

Lime is applied to the soil either in the form of the oxide or carbonate. Sometimes one form is preferred, but often it is considered immaterial which is used, except as economic considerations may dictate. It is known that quicklime may sometimes have an injurious alkalinity soon after its application, but the assumption is generally made that carbonation quickly takes place, so that the ultimate effect is the same as though the carbonate had been used in the first place. It seemed profitable to study these changes with more exactitude, by using the hydrogen electrode method (6) for determining the concentration of H-ions in soil suspensions.

Three soils of different types were used, (1) a clay adobe, (2) a sandy loam, (3) a silty clay loam. A sample of beach sand was also included for comparison. Two 20-pound lots of each soil were placed in earthenware jars. Commercial quicklime was added to the soils and thoroughly mixed. The proportions used were 0.07 per cent and 0.28 per cent calculated in terms of pure CaO. The soils were made up to approximately an optimum moisture content and kept in this condition for six months. Samples were taken at intervals over this period and the H-ion concentration determined as described in the previous publication already cited (6). The samples were not dried and loss of CO_2 was avoided so far as possible in making the measurements. All determinations were carried out in a strictly comparable way. H-ion con-

TABLE 1

Effect of different forms of lime on soil reaction

NUMBER OF SOIL	DESCRIPTION OF SOIL	TREATMENT	P_H IMMEDIATELY AFTER TREATMENT	P_H AFTER 4 DAYS	P_H AFTER 7 DAYS	P_H AFTER 18 DAYS	P_H AFTER 26 DAYS	P_H AFTER 46 DAYS	P_H AFTER 80 DAYS	P_H AFTER 187 DAYS	P_H AFTER GROWING BARLEY 10 MONTHS AFTER TREATMENT	P_H SOIL + 0.10 PER CENT $CaCO_3$	P_H SOIL + 0.40 PER CENT $CaCO_3$
1	Clay adobe	None	7.56									7.39	7.39
1	Clay adobe	0.07 per cent CaO	8.56	8.36	8.56	8.00	8.50	8.05	8.74	8.00	8.22		
1	Clay adobe	0.28 per cent CaO.	10.16	9.26	9.43	9.08	8.74	8.94	9.00	8.70	8.84		
2	Sandy loam	None	7.56									7.87	7.80
2	Sandy loam	0.07 per cent CaO	9.25	8.18	8.50	8.05	8.25	8.11	8.56	8.29	7.87		
2	Sandy loam	0.28 per cent CaO	10.96	10.00	9.85	9.05	9.05	8.94	9.00	8.67	8.43		
3	Silty clay loam	None	7.77									7.80	7.81
3	Silty clay loam	0.07 per cent CaO	9.05	8.43	8.63	8.36	8.29	8.15	8.15	8.15	7.80		
3	Silty clay loam	0.28 per cent CaO	10.30	9.43	9.47	8.84	8.94	8.88	8.91	8.50	8.32		
4	Beach sand	None	7.35									8.08	8.18
4	Beach sand	0.07 per cent CaO	10.82	9.00	9.43	8.53	8.53	8.32	8.29	7.92			
4	Beach sand	0.28 per cent CaO.	12.45	11.68	11.51	10.89	10.65	9.61	9.05	8.39			

centrations are expressed as exponents, 7.10 being taken as neutrality, since it represents the H-ion concentration of pure water. The larger the exponent the greater is the intensity of alkalinity.

The results in table 1 indicate that immediately after the addition of CaO a very considerable increase in OH-ion concentration has been produced. With the larger application the increase is much greater than with the smaller quantity. Comparing the different soils, we find that the reaction of the clay adobe is least affected, while that of the sand undergoes the most marked change. Within three days there is a very great decrease in alkalinity in all cases. Thereafter the decrease is slower and six months later the treated soils still show an increased alkalinity due to CaO. The same soils when treated with equivalent or greater quantities of $CaCO_3$ do not have their reaction appreciably changed. All of the soils were slightly alkaline originally, but similar effects would probably be observed with acid soils, once an excess of CaO were added over that necessary to neutralize the acids present. In connection with the use of $CaCO_3$, Plummer (5) has recently shown that certain soils, to which a very large excess of $CaCO_3$ had been added, a number of years after treatment showed a decidedly high concentration of OH-ion. The carbonate seems to have reacted with the soil constituents, possibly forming silicates.

As a further test of the effect of CaO, a preliminary vegetation experiment was made with barley. Soils 1, 2 and 3 were used after contact with CaO for six months as previously indicated. After harvesting the crop the reaction of the soils was determined again and it was found that no significant change in the OH-ion concentration had occurred. The alkalinity was still greater than that of the untreated soil, but no inhibition of plant growth could be observed. In this connection it should be remembered that any excretion of CO_2 by the plant roots would tend to decrease the alkalinity, at least in the adjacent region.

In the course of another experiment two tanks of an acid fine sandy loam were treated with an excess of lime (about 0.5 per cent CaO), using on one lot CaO and on the other an equivalent quantity of $CaCO_3$. After standing out-of-doors for one year, samples were taken from both tanks and the H-ion concentration determined. The soil treated with $CaCO_3$ gave an exponent of 8.05, while that treated with CaO gave 11.34. The latter represents an intense alkalinity and it is clear that complete carbonation did not take place even after a long period of time. Several tests were made on the nitrifying power of the soil, 0.2 per cent $(NH_4)_2SO_4$ being used. The soil treated with $CaCO_3$ gave a nitrification of 100 per cent. In the case of the CaO-treated soil, no nitrification was observed after four weeks.

Hutchinson and MacLennan (3, 4) have made extensive studies of the effects of CaO and $CaCO_3$ in producing a partial sterilization of the soil. They found that $CaCO_3$ did not produce such sterilization but that CaO was capable of accomplishing this result. The quantity of lime required seemed to depend

upon the type of soil. These observations accord with the measurements of OH-ion concentration just described. In some soils there is considerable immediate chemical reaction between the CaO and some of the soil constituents, so that the OH-ion concentration may never be excessive unless a large percentage of CaO is used. On the other hand, even a small percentage in other soils may cause initially a very high concentration of OH-ions. The maximum effect is noted in sand. After the first few days the alkalinity decreases in all soils, either because of continued chemical action or of slow reaction with CO_2.

In summarizing the present experiments we may say that CaO when first added to a soil produces a high concentration of OH-ion. This decreases markedly within a few days and continues to decrease slowly over a considerable period, but the treated soils still show a greater intensity of alkalinity even after many months, as compared with the untreated soils or those treated with $CaCO_3$. When a considerable excess of CaO is used it is possible for certain soils to maintain such a high concentration of OH-ion that nitrification is practically inhibited. Thus the ultimate effects of CaO and $CaCO_3$ may not necessarily be identical. It is highly desirable that more exact observations be made of the reactions (H-ion concentration) produced by lime in order that its effects, chemical and biological, may be compared with the measurements of this important and definite factor.

SUMMARY

1. The hydrogen-ion concentration of several types of soil was determined after the addition of CaO and $CaCO_3$.

2. The initial effect of both low and high percentages of CaO was to greatly increase the OH-ion concentration. This decreases with time but is higher than in the case of the untreated soil even after ten months. These results are discussed with reference to bacterial activity and partial sterilization of the soil.

REFERENCES

(1) Burd, John S. 1918 Water extractions of soils as criteria of their crop producing power. *In* Jour. Agr. Res., v. 12, no. 6, p. 297–309.

(2) Hoagland, D. R. 1918 The freezing point method as an index of variations in the soil solution due to season and crop growth. *In* Jour. Agr. Res., v. 12, no. 6, p. 369–395.

(3) Hutchinson, H. B. 1912 The action of quicklime on the soil. *In* Rpt. Brit. Assoc. Adv. Sci., 1912, p. 740.

(4) Hutchinson, H. B., and MacLennan, K. 1914 The relative effect of lime as oxide and carbonate on certain soils. *In* Jour. Agr. Sci., v. 6, no. 3, p. 302–322.

(5) Plummer, J. K. 1918 Studies in soil reaction as indicated by the hydrogen electrode. *In* Jour. Agr. Res., v. 12, no. 1, p. 19–31.

(6) Sharp, L. T., and Hoagland, D. R. 1916 Acidity and adsorption in soils as measured by the hydrogen electrode. *In* Jour. Agr. Res., v. 7, no. 3, p. 123–145.

(7) Stewart, G. R. 1918 Effect of season and crop growth in modifying the soil extract. *In* Jour. Agr. Res., v. 12, no. 6, p. 311–368.

THE CHEMICAL EFFECTS OF CaO AND $CaCO_3$ ON THE SOIL

PART II. THE EFFECT ON WATER-SOLUBLE NUTRIENTS IN SOILS

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It is a well-known fact that the physical and biological conditions of soils are usually improved by the addition of calcium compounds. The preceding article by Hoagland and Christie (8) described the effect of different forms and amounts of lime on the reaction of typical soils. This paper shows that the initially high alkalinity produced by calcium oxide is rapidly reduced and that this effect is most pronounced in heavy soils. It seems probable, therefore, that this decrease in alkalinity is not entirely due to carbonation but at least in part to chemical reaction with the soil. In the case of calcium carbonate McIntire (11) states that when this compound is applied to soils in excess of the amount necessary for neutralization, the residual carbonate is gradually decreased, demonstrating "the existence of a long continued reaction between soils and carbonates."

The prevailing idea has been that lime compounds render certain other elements such as magnesium, potassium and phosphates more available, due to the phenomenon of interchange of bases. However, certain experiments seem to refute this theory. Lyon and Bizzell (9) found no increase of potassium in the drainage water or crops of limed soils, although magnesium was increased. The same authors in another paper (10) report the increase of soluble sulfates in limed soils. Bradley (3) found that gypsum acted as an indirect potassium fertilizer, while lime did not and that neither affected the water-soluble phosphates in soils. Briggs and Breazeale (4) have recently shown that pegmatite and orthoclase as well as soils containing these minerals do not yield increased water-soluble potassium when shaken with an aqueous solution of calcium hydrate or calcium sulfate. On the other hand, Andre (1) states that glauconite yields part of its potassium when treated with calcium carbonate. Morse and Curry (12) found that lime and gypsum increased the solubility of potassium in feldspar, claiming that this effect may not be observed when soils are similarly treated, owing to the removal of potassium from solution by the absorbent power of clay. Gaither (6) concludes that lime increases the availability of phosphates by replacement of iron and aluminum. He shows distinctly that silicates are decomposed but states that the assimilation of potassium by crops was decreased by caustic lime.

TABLE 1

Effects of CaO *and* $CaCO_3$ *on water extracts and osmotic pressures of soils*

		NO. 1 SANDY LOAM LEACHED	NO. 2 FINE SANDY LOAM STORED	NO. 3 FINE SANDY LOAM STORED	NO. 4 SILTY CLAY LOAM CROPPED	NO. 5 SILTY CLAY LOAM STORED	NO. 6A SILTY CLAY LOAM CROPPED	NO. 6B SAME AS 6A FALLOWED	NO. 7A CLAY ADOBE CROPPED	NO. 7B SAME AS 7A FALLOWED
Osmotic pressure (atm)	Untreated	0.43	4.94	3.13	0.59	1.93	0.78	5.06	0.69	1.46
		0.46	5.18	3.13	0.64	1.80	0.81	5.18	0.80	1.64
	$CaCO_3$	0.51	5.30	3.25	0.87	1.80		4.82	0.82	1.53
		0.49	5.42	3.25	0.75	1.80		5.18	0.75	1.69
	Average per cent increase	12	6	4	31	−3		−2	6	4
	CaO	0.59	5.50	3.37	0.83	1.93	1.05	5.18	1.06	1.53
		0.57	5.56	3.37	0.88	2.05	1.07	5.18	0.88	1.60
	Average per cent increase	29	8	7	39	6	33	1	32	
Total solids (p.p.m.)	Untreated	318	2332	1760	351	838	448	3788	598	1236
		285	2344	1756	369	809	461	3723	652	1359
	$CaCO_3$	342	2393	1870	475	803		3769	763	1352
		311	2346	1766	428	723		3795	753	1320
	Average per cent increase	8	1	4	26	−7		1	21	3
	CaO	371	2322	1885	428	996	559	3729	794	1335
		374	2322	1836	483	975	710	3801	786	1382
	Average per cent increase	23	−1	6	27	19	39		27	4
Calcium (p.p.m. Ca)	Untreated	32	352	174	33	93	53	396	42	135
		30	343	177	33	99	63	389	42	135
	$CaCO_3$	34	357	177	40	96		419	62	171
		38	354	177	33	93		413	81	164
	Average per cent increase	15	2	1	10	−1		6	70	24
	CaO	40	398	226	33	133	82	419	68	164
		35	404	220	36	133	85	440	71	167
	Average per cent increase	20	15	32	2	38	45	9	66	22

Magnesium (p.p.m. Mg)	Untreated	16	3.6	4.4	17	53	42	30	33	39
		16	3.8	4.5	17	54	42	29	32	36
	$CaCO_3$	17	3.7	4.7	27	47		36	33	38
		17	4.0	4.5	29	46		37	36	39
	Average per cent increase	5	5	2	66	−13		24	7	2
	CaO	19	3.7	4.4	31	47	40	38	38	39
		19	3.7	4.4	33	48	43	38	38	38
	Average per cent increase	16		−2	92	−11	−2	30	16	2
Potassium (p.p.m. K)	Untreated	24	53	128	20	65	38	123	44	59
		24	52	127	20	62	42	123	49	57
	$CaCO_3$	28	56	132	31	49		119	50	46
		29	60	124	32	52		123	49	48
	Average per cent increase	18	10		58	−21		−2	6	−19
	CaO	35	48	121	41	52	43	118	42	44
		37	50	110	38	51	41	121	44	48
	Average per cent increase	50	−5	−10	97	−19	7	−2	−8	−20
Phosphate (p.p.m. PO_4)	Untreated	7.9	7.0	39	5.8	22	7.9	3.3	13	7.3
		7.5	6.4	37	5.5	23	7.2	4.6	14	5.6
	$CaCO_3$	9.6	5.3	37	10.8	25		4.0	12	3.2
		9.6	7.0	38	11.4	23		4.0	10	2.2
	Average per cent increase	25	−7	−2	95	6			−18	−60
	CaO	9.0	8.2	33	10.1	22	7.2	4.6	11	7.1
		11.3	8.2	32	9.1	27	8.5	7.9	14	11.2
	Average per cent increase	33	22	−15	69	10	4	57	−3	40
Sulfate (p.p.m. SO_4)	Untreated	4.3	175	151	17	83	58	258	108	122
		5.4	180	147	19	86	57	247	107	119
	$CaCO_3$	28.7	171	146	27	84		302	125	115
		28.6	176	146	44	82		297	129	121
	Average per cent increase	485	−2	−2	44	−2		18	19	−3
	CaO	23.6	171	128	24	73	45	297	120	132
		23.6	197	142	24	70	47	288	120	130
	Average per cent increase	382	3	−9	35	−15	−20	15	12	9

TABLE 1—(Concluded)

		NO. 1 SANDY LOAM LEACHED	NO. 2 FINE SANDY LOAM STORED	NO. 3 FINE SANDY LOAD STORED	NO. 4 SILTY CLAY LOAM CROPPED	NO. 5 SILTY CLAY LOAM STORED	NO. A6 SILTY CLAY LOAM CROPPED	NO. 6B SAME AS 6A FALLOWED	NO. 7A CLAY ADOBE CROPPED	NO. 7B SAME AS 7A FALLOWED
Nitrate (p.p.m. NO_3)	Untreated	20	867	507	53	153	40	1036	12	319
		19	920	502	50	152	38	1035	12	340
	$CaCO_3$	20	901	524	52	159		1056	25	319
		21	866	530	54	152		1036	22	340
	Average per cent increase	3	−1	4	2	2		1	102	
	CaO	19	885	531	52	153	42	1076	16	324
		20	918	525	55	152	41	1058	12	329
	Average per cent increase		1	4	4		6	3	18	

It is evident from a consideration of these and other references that there is considerable divergence of opinion concerning the effect of calcium compounds on the available nutrients in soils. The purpose of the present paper is to discover possible reasons for this apparent disagreement between the several investigations on the important question of the indirect fertilizing action of lime compounds.

During an extensive investigation of the water extracts of soils carried on by this laboratory (5,13), the methods of analysis suitable for such extracts have been considerably improved and are believed to permit of more exact interpretation than was formerly the case. The present paper reports the data obtained from a study of the effects of calcium oxide and calcium carbonate on the concentration of the soil solution as measured by the water-extraction and freezing-point methods (2).

DESCRIPTION OF SOILS

None of the soils used in this investigation were acid and therefore no portion of the lime applied was used for neutralization. In table 1 is given the type and previous treatment of each soil. It should be noted that sandy loam no. 1, silty clay loams no. 4 and no. 6a and clay adobe no. 7a contained relatively low amounts of water-soluble nutrients, due in the case of the sandy loam to excessive leaching and in the others to the depletions caused by several seasons' cropping. The remaining soils which had been stored in bins or fallowed for several years showed a considerably greater accumulation of soluble plant-food. This is in accord with the recent findings of Stewart (13) who has shown that stored or fallowed soils show greater concentrations of all nutrients save phosphates than the same soil under crop. Soils 4, 5 and 6, although from different localities, are of the same series, Yolo silty clay loam, according to the soil surveys of the Bureau of Soils of the United States Department of Agriculture. This will be referred to later in connection with the different effects of lime on these soils.

PROCEDURE

Six 500-gm. portions of each air-dry soil were weighed into quart jars. To two jars of each soil was added 0.5 gm. C. P. $CaCO_3$ (equal to an application of 2 tons per acre-foot of 4,000,000 pounds), which was mixed with the soil before moistening. The next two jars were treated with 0.28 gm. C. P. CaO (equivalent in amount of calcium to the $CaCO_3$ application), added in a water suspension. This was found to be the most efficient method of mixing the oxide with the soil. The remaining two jars were not limed and served as controls. Certain investigators have used excessive quantities of lime in similar studies. In order to make the results of practical value, the amounts of lime here used are comparable with moderate field applications. After the addition of the calcium compounds, sufficient distilled water was added

to all jars to bring the soil to optimum, following which the contents of each jar were again stirred. All jars were then allowed to stand one week at ordinary temperatures, being loosely covered to prevent excessive evaporation, yet allowing easy access of air. At the end of seven days, the contents of each jar were removed and mixed. A 100-gm. portion was dried to constant weight to determine moisture. A 340-gm. sample was extracted with five times its weight of distilled water after the manner described by Stewart (13). The remainder was used to determine the osmotic pressure of the soil solution. This was done according to the method of Bouyoucos and McCool (2) which consists in determining the lowering of the freezing point directly in the soil under optimum moisture conditions.

Table 1 gives a complete résumé of the results obtained. The concentration of the soil solution is reported in atmospheres of osmotic pressure. The analyses of the soil extracts are reported as parts per million of water-free soil. Below each pair of duplicate determinations is shown the average per cent of increase or decrease resulting from the CaO and $CaCO_3$ additions.

DISCUSSION OF RESULTS

In any chemical study of soil fertility it is essential to determine the probable variation inherent in the methods used. For a complete discussion of the methods of extraction and analysis used in this investigation the reader is referred to the recent work of Stewart (13). For present purposes it is sufficient to state that the maximum variation between duplicate extractions of the same soil, for any element, was found to be 10 per cent and that the mean factor of error is approximately 3 per cent. The usual close agreement between duplicates shows that large variations are the exception rather than the rule. No conclusions should be drawn except where the increase or decrease due to lime is well above the average error.

Concentration of soil solution. Soils 1, 4, 6a and 7a show that the concentration of the soil solution has been materially increased by the addition of lime, the effect of CaO being more pronounced than that of $CaCO_3$. The remaining soils show little or no effect. It is interesting to note here that the soils showing increases are those recently cropped or having a low concentration, while those showing no effect contain a much greater concentration of nutrients due to fallowing or storage.

Total solids. Hoagland (7) has shown that although the 1:5 water extraction removes more material than is actually in the soil solution, there is a distinct correlation between these two values. In this connection it should be noted that the soils showing substantial increases of total solids on extraction are the same soils which show an increased concentration of the soil solution and the percentage increases are of approximately the same magnitude for both determinations.

Calcium. The increases in the case of calcium are quite variable, the CaO on the whole giving more soluble calcium than the $CaCO_3$. The manner in which the different soils react with lime compounds is reflected by the variable increases in water-soluble calcium.

Magnesium. The cropped soils, nos. 1, 4 and 7a, show increases in soluble magnesium. In the case of soil 6 this tendency is reversed, the fallowed soil giving an increase and the cropped soil none. Soil 4 shows approximately a 100 per cent increase in available magnesium which will be discussed later in connection with its possible effect on the lime-magnesia ratio. Soil 5 shows a distinct depression of soluble magnesium.

Potassium. Soils 1 and 4 unquestionably show large increases of available potassium due to both CaO and $CaCO_3$, CaO being the more potent. These two soils were the lowest in soluble potassium before treatment. The remaining soils show no increase, and in fact soils 5 and 7b experience a considerable depression of soluble potassium due to the lime. An inspection of the results obtained for this important element points out a probable reason for the disagreement between various investigators concerning the effect of lime as an indirect potash fertilizer.

Phosphate. Owing to the difficulty of obtaining closely agreeing duplicates in all cases, as well as to the small number of parts per million usually involved, no great confidence is felt in the results for this determination. However, soils 1 and 4 show marked increases in available phosphate for both CaO and $CaCO_3$ treatments, which increases are believed to be significant. The remaining data are not sufficiently in agreement to warrant discussion here. It might be conceived that the increased alkalinity due to lime may repress phosphate solubility but little or no evidence in favor of this theory is noticed here.

Sulfate The three cropped soils, nos. 1, 4 and 7a, and also 6b, produce increases in soluble sulfates. This is especially marked in soil 1 which shows an increase of approximately 400 per cent for both CaO and $CaCO_3$ treatments. The two soils showing the greatest increases are those lowest in soluble sulfates.

Nitrates. With the single exception of soil 7a, no soils show significant increases in nitrates. The small absolute increase of nitrates in this soil produces a large percentage increase, because of the low amount of nitrates resulting from crop depletion. It will be remembered that these soils had been allowed to stand only one week at ordinary temperatures. If nitrification may be taken as a partial index of bacterial activity in the soil, then the observed effects of lime on these soils must be due to a purely chemical reaction and not to any stimulation of the biological agencies in the soil.

GENERAL DISCUSSION

In order to demonstrate more conclusively the direct chemical effect of the lime, another sample of soil 4 was treated with 0.4 per cent CaO (8 tons per

acre-foot) and allowed to stand only 24 hours before analysis. The following results unquestionably show the immediate chemical effect of the quicklime.

	Total solids.	Ca	Mg	K	PO_4	NO_3
	p.p.m.	*p.p.m.*	*p.p.m.*	*p.p.m.*	*p.p.m.*	*p.p.m.*
Limed	274	34	13	32	5	19
Not limed	505	64	23	46	9	14

Larger portions of soils 1, 4 and 7a, in gallon crocks, after being treated with CaO and $CaCO_3$, were planted to barley. The yield of barley in the limed soils reflected the beneficial effects of the calcium, being from two to three times greater than the controls. This increased yield may be largely accounted for by improved physical and biological conditions. Without giving the detailed figures, it may be stated that the analyses of the soils before planting showed marked increases in total solids, calcium, magnesium and potassium, and it is believed that these increases in available nutrients must account, at least in part, for the increased crop.

It is a well-known fact that lime decomposes organic matter in soils and therefore it might well be contended that the increases in total solids were largely due to soluble organic matter. In the case of about half the soils examined, the electrical conductivity of the soil extract was measured. The figures obtained were in direct relation to the total solids determination, showing that at least the greater part of the increases were due to inorganic and not to organic matter.

Certain other inorganic elements, such as sodium, silicon, etc., have not been determined. Where increases in the nutritive elements do not account for the total increase in total solids, it is probable that such other undetermined elements have been made more soluble. Gaither (6) has previously shown the decomposition of silicates in soils by lime. It would be perfectly reasonable to assume that other elements such as silicon and sodium have been rendered more soluble in some of the soils.

Soils 5 and 6b are of the same type and series and yet react with lime in different fashion. This indicates that the changes induced by liming are more deep-seated than can be accounted for by soil classification.

Lyon and Bizzell (9) found in a certain soil that the addition of lime caused a narrower lime-magnesia ratio in the drainage water. If there is any value in a certain lime-magnesia ratio for soils, it must be concerned with the water-soluble elements. An inspection of the figures for calcium and magnesium in soil 4 shows that while the original soil had a lime-magnesia ratio of 2:1, after liming the ratio became 1:1, as a result of the liberation of magnesium by the calcium. Certain of the other soils produced a broader ratio due to increases in calcium or depressions in magnesium, or both. It is seen from this that the calcium-magnesium ratio of a soil solution cannot be predicted prior to the addition of lime.

It is evident from the data considered that all soils do not react chemically with lime in the same manner. Furthermore, even the same soil under different concentrations of soil solution, due to cropping or fallowing, may react differently. No attempt is made to explain the reasons for the changes observed. Evidently, the addition of lime compounds changes the equilibrium in the complex soil solution and the resultant effects are varied and impossible to predict. Further study involving determinations of all the elements concerned would be necessary before even a plausible hypothesis could be advanced.

SUMMARY

1. The direct chemical effects of CaO and $CaCO_3$ on the water-soluble nutrients in 7 soils of 3 different types have been studied. Supplementary data by freezing-point and conductivity methods have been obtained.

2. CaO and $CaCO_3$ increased the water-soluble potassium in only 2 soils, and in 2 soils caused a depression. Soluble magnesium was increased in 4 soils, and decreased in 1 case. Soluble sulfates were increased in 4 soils. Only 2 soils showed a significant increase in phosphates.

The effects of CaO are more pronounced than those of $CaCO_3$.

3. The effect of lime on soils depends in part upon the previous treatment of the soil, particularly with respect to the concentration of the soil solution. A greater effect was observed in soils having a low initial concentration.

REFERENCES

(1) ANDRÈ, G. 1916 Displacement of the potash and phosphoric acid contents of certain rocks by some fertilizers. *In* Compt. Rend. Acad. Sci. [Paris], t. 162, no. 3, p. 133–136.

(2) BOUYOUCOS, G. J., AND MCCOOL, M. M. 1915 The freezing point method as a new means of measuring the concentration of the soil solution directly in the soil. Mich. Agr. Exp. Sta. Tech. Bul. 24.

(3) BRADLEY, C. E. 1910 Reaction of lime and gypsum on some Oregon soils. *In* Jour. Indus. Engin. Chem., v. 2, no. 12, p. 529–530.

(4) BRIGGS, L. J., AND BREAZEALE, J. F. 1917 Availability of potash in certain orthoclase bearing soils as affected by lime or gypsum. *In* Jour. Agr. Res., v. 8, no. 1, p. 21–28

(5) CHRISTIE, A. W., AND MARTIN, J. C. 1917 The volumetric determination of sulfates in water extracts of soils. *In* Soil Sci., v. 4, no. 6, p. 477–479.

(6) GAITHER, E. W. 1910 Effect of lime upon the solubility of soil constituents. *In* Jour. Indus. Engin. Chem., v. 2, no. 7, p. 315–316.

(7) HOAGLAND, D. R. 1918 The freezing point method as an index of variations in the soil solution due to season and crop growth. *In* Jour. Agr. Res., v. 12, no. 6, p. 369–395.

(8) HOAGLAND, D. R., AND CHRISTIE, A. W. 1918 The chemical effects of CaO and $CaCO_3$ on the soil. Part I. The effect on soil reaction. *In* Soil Sci., v. 5, p. 379–382.

(9) LYON, T. L., AND BIZZELL, J. A. 1916 Calcium, magnesium, potassium and sodium in the drainage water from limed and unlimed soil. *In* Jour. Amer. Soc. Agron., v. 8, no. 2, p. 81–87.

(10) LYON, T. L., AND BIZZELL, J. A. 1916 The loss of sulfur in drainage water. *In* Jour. Amer. Soc. Agron., v. 8, no. 2, p. 88–91.

(11) MCINTIRE, W. H. 1916 Factors influencing the lime and magnesia requirements of soils. Tenn. Agr. Exp. Sta. Bul. 115.

(12) MORSE, F. W., AND CURRY, B. E. 1909 The availability of soil potash in clay and clay loam soils. N. H. Agr. Exp. Sta. Bul. 142, p. 39–58.

(13) STEWART, G. R. 1918 Effect of season and crop growth in modifying the soil extract. *In* Jour. Agr. Res., v. 12, no. 6, p. 311–368.

THE RELATIVE "RAWNESS" OF SOME HUMID SUBSOILS[1]

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INTRODUCTION

Three recent articles (1, 2, 4) dealing with the relative "rawness," or unproductivity, of subsoils as compared with surface soils make it appear that the prevailing views upon the subject rest upon an uncertain foundation. The unproductivity of the subsoils of humid regions has long been recognized, as Alway, McDole and Rost (2, p. 9–10) have pointed out, and no distinction has been made between non-leguminous plants and inoculated legumes. On the contrary, the subsoils of arid regions have been regarded as characterized by a lack of any such rawness, Hilgard's views upon these having been accepted without question.

Alway, McDole and Rost presented evidence from both pot experiments and field observations in support of the view that certain humid subsoils, viz., those developed on the loess of eastern Nebraska, with inoculated legumes are practically as productive as the corresponding surface soils, although very unproductive with non-leguminous plants.

Lipman (4) denies the correctness of Hilgard's views upon the rawness of arid subsoils in so far as these concern non-legumes, and concludes from his own observations in California that the subsoils of arid regions are practically as raw toward such plants as are the typical subsoils of humid regions. He further questions the actual existence of any proof of the rawness of humid subsoils toward legumes, or, as he puts it, whether legumes will not grow on these. But, as Alway has emphasized (1, p. 197), the question of rawness depends not upon whether the plants will merely *grow* but upon whether they will *thrive*, or make a growth at all comparable with that on the corresponding surface soils.

The experiments described below, undertaken at the suggestion of Dr. F. J Alway and carried out as subsidiary to a chemical study of prairie soils in comparison with those still covered with virgin forest, furnish evidence of as great a productivity in the case of some of the forest subsoils as that of the subsoils of the prairie-covered loess reported by Alway, McDole and Rost, while the corresponding prairie subsoils, with fewer of the characteristics typical

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of those of humid regions, were found to be decidedly unproductive. As the author's work on the subject has now been interrupted for an indefinite period the data so far accumulated are presented below.

PROPERTIES OF THE SOILS

The location of the fields from which the soils were secured and the differences in precipitation and natural vegetation are shown in figure 1. In the case of

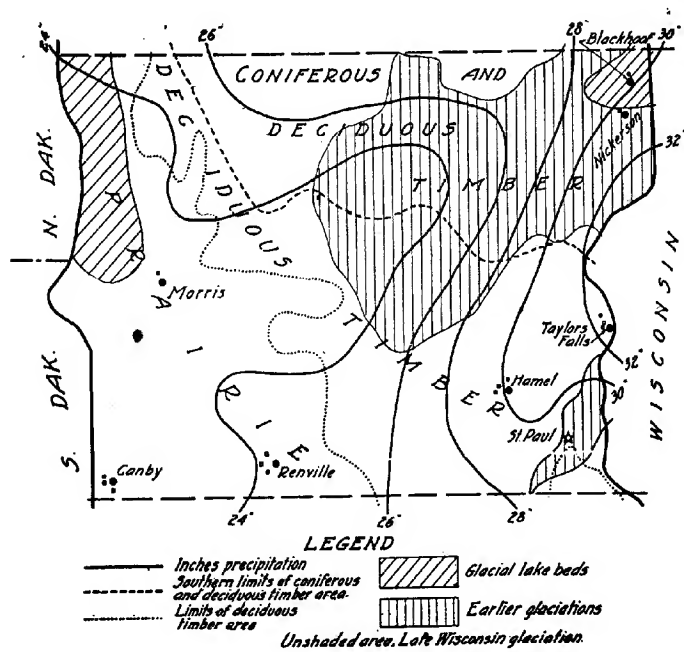


FIG. 1. MAP OF PORTION OF MINNESOTA

Showing sources of soils used, boundaries of the Late Wisconsin glaciation (after Leverett, 3, p. 32), normal precipitation (after Purssell, 3, p. 22), and distribution of forest and prairie (after Upham and Butters, 3, p. 39).

all except those from Blackhoof and Nickerson, both surface soil and subsoil consist of till from the Late Wisconsin glaciation. The Blackhoof soils are of lacustral origin, having been deposited in glacial Lake Duluth (3, p. 32) and the Nickerson samples are from a very sandy moraine at the edge of this lake. Accordingly, all except those from Nickerson are products of the very latest continental ice sheet and so have been exposed to the action of the weather and of vegetation for approximately the same length of time.

In table 1 are indicated the important properties of the different soils used in the vegetation experiments. The organic carbon was determined by com-

bustion with copper oxide in a current of oxygen. The degree of acidity reported is that found by the Truog method. Effervescence was tested by treating the thoroughly moist sample with 10 per cent hydrochloric acid. It will be seen that in the case of a few soils the reaction became less acid during the course of the experiment, as the result of the use of tap water which con-

TABLE 1

Composition and properties of the soils used

LOCALITY	DEPTH	MOISTURE EQUIVALENT	NITROGEN	ORGANIC CARBON	ORGANIC MATTER*	REACTION OF SOIL		EFFERVESCENCE WITH HCl
						At beginning	At conclusion	
			per cent	*per cent*	*per cent*			
Canby.....	1 to 6 in.	30.25	0.329	4.015	6.922	Neutral	Neutral	None
	3rd ft.	23.24	0.051	0.770	1.328	Neutral	Neutral	Strong
Morris.....	1 to 6 in.	30.46	0.330	4.444	7.662	Neutral	Neutral	None
	1st ft.	27.78	0.280	3.303	5.695	Neutral	Neutral	None
	3rd ft.	18.16	0.042	0.642	1.107	Neutral	Neutral	Strong
	5th ft.	17.39	0.013	0.244	0.421	Neutral	Neutral	Strong
Renville...	1 to 6 in.	35.91	0.393	5.134	8.852	Neutral	Neutral	None
	3rd ft.	27.28	0.051	0.865	1.491	Neutral	Neutral	Strong
Hamel.....	1 to 6 in.	23.02	0.181	2.405	4.146	Slightly acid	Very slightly acid	None
	3rd ft.	26.72	0.048	0.390	0.672	Neutral	Neutral	Very slight
Taylors Falls.	1 to 6 in.	19.47	0.152	1.900	3.276	Slightly acid	Slightly acid	None
	3rd ft.	18.12	0.029	0.248	0.428	Slightly acid	Neutral	None
Blackhoof	1 to 6 in.	35.13	0.188	2.732	4.710	Medium acid	Medium acid	None
	3rd ft.	41.91	0.026	0.295	0.509	Neutral	Neutral	Strong
	5th ft.	52.50	0.022	0.345	0.595	Neutral	Neutral	Strong
Nickerson	1 to 6 in.	6.95	0.068	0.951	1.640	Strongly acid	Medium acid	None
	3rd ft.	1.56	0.006	0.094	0.162	Very slightly acid	Neutral	None

* Computed from organic carbon. Organic matter = C x 1.724

tained a considerable amount of calcium bicarbonate. As a single-valued expression of the texture the moisture equivalent is reported.

Three sets of soils, those from Morris, Blackhoof and Nickerson, were taken from a single field each. The samples from Morris were composites from 20 pits 2 rods or more apart, and those from Blackhoof from 3 pits 5 rods apart, while those from Nickerson were from 10 pits 16 rods or more apart. The sets from the four other localities were each composites from 3 fields, 2

miles or more apart. In each of these latter fields, composite samples were prepared from 5 pits 2 rods or more apart; the same volume of soil from each pit was used in making the field composites and the same weight of each of these field composites in making the area composites used in the vegetation experiments.

None of the fields had ever been brought under the plow, those at Canby, Renville and Morris being prairie fields used as meadows, while those at Hamel and Taylors Falls formed remnants of the original deciduous forest, few trees having been cut and no sign of injury by fire being found. The original forest of red and jack pine at Nickerson and of white pine at Blackhoof had been cut off by the lumbermen and both areas had, some twenty-four years ago, been burned over by severe forest fires. A young growth, chiefly deciduous, had since sprung up.

The bulk soils after reaching the laboratory were exposed to the air long enough to make them almost air-dry, then thoroughly mixed and tamped into the vegetation vessels.

EXPERIMENT I. SOILS FROM THREE LEVELS

In the first experiment, 1915–1916, soils from only Morris and Blackhoof were employed, each being represented by three samples, viz., the surface 6 inches and the third and fifth feet from the latter and the first, third and fifth feet from the former. Wooden boxes, 12 inches square and 8 inches deep, coated on the inside with pitch tar and provided with holes in the bottom for drainage, were employed as vegetation vessels. Four were filled from each sample. The soil was thoroughly moistened and at once planted to corn, but, at the end of two weeks, and before the plants had begun to show any distinct differences, the corn was removed, two of the boxes planted to barley and the other two to alfalfa. All the soils were kept well moistened with tap water.

The barley, early thinned to 12 plants per box, made a normal growth on the surface soil, producing well filled heads, but that on the subsoil was stunted, yellowish-green in color and formed very little seed. The alfalfa, at the end of about two weeks was thinned to 18 plants per box and treated with a suspension prepared by macerating nodule-bearing alfalfa roots in water. A second application insured the inoculation of all the plants, as indicated by their dark green color. Three crops were harvested. The appearance of the plants (plate 1) and the yields (table 2) indicate that the subsoils from both places were quite raw, being practically as unproductive of alfalfa as of barley. The latter was not cut until it was thoroughly ripe, but before the total dry weight and the weights of grain had been determined the plants were accidentally destroyed. The heads of the plants from the subsoils contained very few seeds, thus making the differences between the surface soil and subsoil crops much greater than the number of heads would indicate.

EXPERIMENT II. SOILS FROM TWO LEVELS

In the second experiment only two depths were represented, the surface 6 inches and the third foot. Fresh material from Blackhoof and Morris was employed along with recently collected samples from the other places. The soils, prepared as in the first experiment, were placed in galvanized iron pots, 12 inches deep, with a surface area of 0.75 square foot, and painted on the inside with pitch tar. Drainage was effected by holes in the bottom and a

TABLE 2

Yields of crops in first experiment

SOIL	POT NO.	ALFALFA (DRY MATTER)				BARLEY				
		First crop June 12	Second crop July 19	Third crop Sept. 30	Total 3 crops	Green weight	No. of culms	No. of plants	Average height of plants	No. of heads
Prairie soil from Morris										
		gm.	*gm.*	*gm.*	*gm.*	*gm.*			*inches*	
First foot	1	21.4	13.1	13.4	47.9	72.7	28	12	32.0	21
	2	20.0	15.5	14.0	49.5	67.0	23	11	33.0	15
Average		20.7	14.3	13.7	48.7	69.8	25	11	32.5	18
Third foot	1	3.8	4.0	5.7	13.5	18.3	10	10	19.5	8
	2	4.0	5.1	6.3	15.4	17.0	11	11	19.0	7
Average		3.9	4.5	6.0	14.4	17.6	10	10	19.2	7
Fifth foot	1	1.8	1.7	2.4	5.9	8.3	9	9	16.0	7
	2	2.2	2.4	2.3	6.9	9.5	12	11	15.0	5
Average		2.0	2.0	2.3	6.4	8.9	10	10	15.5	6
Forest soil from Blackhoof										
1 to 6 inches	1	27.8	28.4	19.0	75.2	92.0	41	11	32.0	21
	2	33.1	36.6	23.9	93.6	112.0	38	10	33.0	15
Average		30.4	32.5	21.4	84.4	102.0	40	10	32.5	18
Third foot	1	3.8	3.5	2.6	9.9	12.0	10	10	19.5	8
	2	2.5	2.1	1.8	6.4	10.0	11	10	19.0	7
Average		3.1	2.8	2.2	8.1	11.0	10	10	19.2	7
Fifth foot	1	2.8	2.2	1.6	6.6	9.5	12	12	16.0	7
	2	3.0	2.7	1.6	7.3	9.0	11	12	15.0	5
Average		2.9	2.4	1.6	6.9	9.2	11	12	15.5	6

half-inch layer of coarse gravel. In this experiment, instead of sowing seed, the writer transplanted 10 alfalfa plants secured from a field sown 5 months before, care being taken to select roots of uniform size. As only plants with adhering nodules were employed it appeared unnecessary to take any further measure to insure inoculation and none of the plants at any subsequent time gave evidence of lack of inoculation. The soil was moistened, the roots spread

TABLE 3

Yields of dry matter from four crops of alfalfa, number of plants surviving and weight of roots

SOIL	POT NO.	YIELD OF DRY MATTER: Preliminary crop December 19, 1917	First crop May 14, 1917	Second crop June 25, 1917	Third crop August 18, 1917	Fourth crop October 3, 1917	Total for 4 crops	NUMBER OF SURVIVING PLANTS	WEIGHT OF ROOTS	WEIGHT OF SOIL PER POT
					Canby					
		gm.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*		*gm.*	*kgm.*
1 to 6 inches	1	8.3	10.7	14.4	19.4	6.5	51.0	8	31	14.9
	2	4.9	12.3	15.5	18.0	6.9	52.7	6	24	14.9
Average		6.6	11.5	14.9	18.7	6.7	51.8	7	27.5	
Third foot	1	6.8	6.4	5.5	5.7	0.5	18.1	10	31	22.0
	2	5.5	4.6	7.9	8.1	1.0	21.6	9	29	22.0
Average		6.1	5.5	6.7	6.9	0.7	19.8	9.5	30	
					Morris					
1 to 6 inches	1	3.4	20.0	19.9	26.9	16.2	83.0	6	35	18.8
	2	8.6	18.7	18.3	31.2	17.6	85.8	6	39	18.8
Average		6.0	19.3	19.1	29.0	16.9	84.4	6	37	
Third foot	1	6.5	7.7	6.3	7.5	2.0	23.5	6	25	22.8
	2	6.5	7.4	7.1	7.3	2.0	23.8	6	24	22.8
Average		6.5	7.5	6.7	7.4	2.0	23.6	6	24.5	
					Renville					
1 to 6 inches	1	7.7	14.8	15.2	27.3	13.3	70.6	9	41	16.7
	2	8.0	13.8	14.0	27.8	14.5	70.1	7	37	16.7
Average		7.8	14.3	14.6	27.5	13.9	70.3	8	39	
Third foot	1	8.0	3.0	6.7	3.8	0.6	14.1	8	28	21.1
	2	4.6	4.8	6.8	4.2	0.7	16.5	9	20	21.1
Average		6.3	3.9	6.7	4.0	0.6	15.3	8.5	24	
					Hamel					
1 to 6 inches	1	4.9	20.8	22.4	25.9	15.4	84.5	7	39	20.5
	2	5.1	19.1	19.3	24.7	12.3	75.4	2	26	20.5
Average		5.0	19.9	20.8	25.3	13.8	79.9	4.5	32.5	
Third foot	1	8.1	16.9	19.0	22.9	12.7	71.5	10	42	21.4
	2	6.6	21.7	20.3	22.9	16.0	80.9	7	36	21.4
Average		7.3	19.3	19.6	22.9	14.3	76.2	8.5	39	

TABLE 3—Continued

SOIL	POT NO.	YIELD OF DRY MATTER: Preliminary crop December 19, 1917	First crop May 14, 1917	Second crop June 25, 1917	Third crop August 18, 1917	Fourth crop October 3, 1917	Total for 4 crops	NUMBER OF SURVIVING PLANTS	WEIGHT OF ROOTS	WEIGHT OF SOIL PER POT
Taylors Falls										
		gm.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*		*gm.*	*kgm.*
1 to 6 inches	1	4.6	20.9	17.0	20.2	13.5	71.6	6	37	21.9
	2	7.0	22.5	18.5	24.6	14.1	79.7	7	41	21.9
Average		5.8	21.7	17.7	22.4	13.8	75.6	6.5	39	
Third foot	1	6.8	24.5	20.5	21.9	15.2	82.1	10	36	22.4
	2	6.3	20.0	18.3	25.6	14.8	78.7	10	38	22.4
Average		6.5	22.2	19.4	23.7	15.0	80.4	10	37	
Blackhoof										
1 to 6 inches	1	6.9	24.1	19.4	32.0	19.2	94.7	7	48	19.0
	2	7.2	25.2	26.8	32.9	22.2	107.1	4	43	19.0
Average		7.0	24.6	23.1	32.4	20.7	100.9	5.5	45.5	
Third foot	1	6.6	2.4	6.1	4.7	1.1	14.3	9	18	18.6
	2	6.0	4.9	6.0	7.2	2.2	20.3	6	20	18.6
Average		6.3	3.6	6.0	5.9	1.6	17.3	7.5	19	
Nickerson										
1 to 6 inches	1	5.9	24.5	17.3	21.8	13.3	76.9	5	37	22.9
	2	4.3	28.4	23.3	19.2	11.9	82.8	3	25	22.9
Average		5.1	26.4	20.3	20.5	12.6	79.8	4	31	
Third foot	1	6.6	13.7	10.3	12.5	5.1	41.6	6	28	26.8
	2	5.4	12.7	12.2	13.3	5.7	43.9	10	28	26.8
Average		6.0	13.2	11.2	12.9	5.4	42.7	8	28	

out as much as possible, the soil pressed firmly about them and more water at once added. By the use of this method it was hoped that a crop would be obtainable in a shorter time and it also promised to assure the penetration of the fine-textured subsoils by the roots, which it was suspected might have partly caused the poor yields of alfalfa in the first experiment.

The transplanting was done on November 12, 1916, and the first crop developed very rapidly during the following two weeks, doubtless as a result of the supply of nutrients in the transplanted roots. At the end of five weeks, on December 19, the growth had practically ceased, and the tops were removed, a remarkably uniform yield from all the soils being shown (table 3). After this first, or preliminary crop, the plants made little growth for several months, the next cutting being secured on May 14. Three additional crops

were obtained, harvested on July 25, August 18 and October 3. The preliminary crop is not included in the total yields, as this is not to be attributed to nutrients derived from the respective soils. After harvesting the last crop the roots were carefully removed from the pots, washed to free them from adhering soil, dried and weighed. In all pots the roots were well scattered throughout the soil mass, penetrating to the bottom and into the washed gravel placed there to facilitate drainage.

The yields from the duplicate pots were very similar, in fact so nearly alike both in the different crops and in the total for all four that the use of single pots of each soil would have led to exactly the same conclusions as to their relative productivity. The content of nitrogen and that of organic matter in the productive subsoils from the forest fields were rather lower than those of the prairie subsoils which proved raw.

TABLE 4

Dry matter in the tops from four crops of alfalfa and in the roots at the end of the experiments

SOIL	TOPS		PRODUCTIVITY OF SUBSOIL	ROOTS		RATIO OF TOPS TO ROOTS	
	Surface	Subsoil	Yield on surface soil = 100	Surface	Subsoil	Surface	Subsoil
	gm.	*gm.*		*gm.*	*gm.*		
Canby	51.8	19.8	38	27.5	30.0	1.9	0.7
Morris	84.4	23.6	28	37.0	24.5	2.3	1.0
Renville	70.3	15.3	22	39.0	24.0	1.8	0.6
Hamel	79.9	76.2	95	32.5	39.0	2.5	2.0
Taylors Falls	75.6	80.4	106	39.0	37.0	1.9	2.2
Blackhoof	100.9	17.3	17	45.5	19.0	2.2	0.9
Nickerson	79.8	42.7	54	31.0	28.0	2.6	1.5

In four of the seven sets, those from Morris, Canby, Renville and Black hoof, the yield on the subsoil was only from one-sixth to two-fifths of that on the corresponding surface soil (table 4). In the Nickerson set the yield on the subsoil was slightly more than half that on the surface material. In the case of the forest-covered fields at Hamel and Taylors Falls the subsoils yielded as well as the corresponding surface soils. Four of the seven subsoils, those from Morris, Canby, Renville and Blackhoof, were very rich in carbonates, all showing a strong effervescence with hydrochloric acid. The fields at the first three places were prairies and the rainfall at all three is lighter than at Hamel or Taylors Falls where the subsoils showed little carbonate.

From table 3 it will be seen that while the number of surviving plants varied from 2 to 10 per pot, the weight of the roots varied but little. Where only a few plants had survived the roots of these showed a much greater development than where few or no plants had died. That this variation in the number of surviving plants exerted no distinct influence upon the yield of alfalfa may be seen by comparing the yields from the duplicate pots, those from Hamel and Blackhoof furnishing especially good illustrations.

The fact that in the case of the four surface soils corresponding to the unproductive subsoils the yields of alfalfa vary with the weight of the soil in the pots, would suggest that the amount of soil to which the roots had access had a much greater influence upon the yields than did the number of plants.

SUMMARY

Alfalfa was grown in the greenhouse on seven sets of Minnesota glacial soils, each set consisting of the surface 6 or 12 inches of virgin soil and the corresponding subsoil taken from the third foot. Three sets were secured from prairie fields with a very calcareous subsoil and the others from forested areas. Two of the subsoils, both low in carbonates and from forest fields, yielded as well as both the corresponding and the other surface soils, while the five other subsoils gave yields only from one-sixth to one-half as high as the corresponding surface soils. The nitrogen content of the productive subsoils was no higher than that of those which proved "raw."

Thus, in the humid state of Minnesota, some of the glacial subsoils are as productive of alfalfa as surface soils when inoculation is assured, but others are quite unproductive, and the rawness is not associated with an especially low nitrogen content or with a lack of carbonates.

REFERENCES

(1) ALWAY, F. J. 1918 The "rawness" of subsoils. *In* Science, v. 47, p. 196–198.

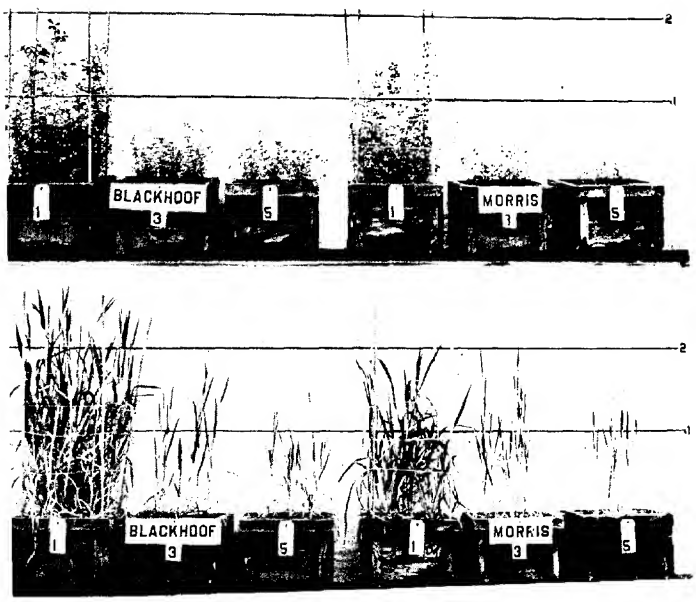
(2) ALWAY, F. J., MCDOLE, G. R., AND ROST, C. O. 1917 The loess soils of the Nebraska portion of the Transition Region: VI. The relative "rawness" of the subsoils. *In* Soil Sci., v. 3, no. 1, p. 9–35.

(3) LEVERETT, F. 1915 Surface formations and agricultural conditions of northwestern Minnesota, with a chapter on climatic conditions of Minnesota by U. G. Purssell. Minn. Geol. Survey Bul. 12, 78 p. and map.

(4) LIPMAN, C. B. 1917 The "rawness" of subsoils. *In* Science, v. 46, p. 288–290.

PLATE 1

Alfalfa and barley on soil from the different foot levels (first, third and fifth), showing the unproductivity of the subsoils toward both crops. In the Blackhoof set the surface sample was from only the first half-foot section.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CHEMICAL CRITERIA, CROP PRODUCTION AND PHYSICAL CLASSIFICATION IN TWO SOIL CLASSES

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Several publications (1, 3, 7) were recently issued from this laboratory embodying the results of an extended series of studies of soils in the cropped and the uncropped condition. The general conclusion presented in these articles was that there is a much closer relationship between the soil solution and water extracts of soils than is commonly supposed. The studies referred to also included a comparison of the results of analysis of all of our soils by the complete (fusion method) hydrochloric acid (sp. gr. 1.115) and citric acid (1 per cent) extraction methods. These latter results were not published inasmuch as they showed little or no relation to crop production and served only to fix our long held conviction that the first two methods at least are absolutely worthless as criteria of the present productivity of soils. Since our work on water extractions appeared, however, we have received a number of requests for information as to the other chemical characteristics of our soils. It therefore seems desirable to publish these latter in order that others may be able to draw their own conclusions as to the relative value of the various methods.

We have been unable to avoid the conclusion that in spite of inconclusive or negative results in the application of the various methods referred to, many workers continue to draw inferences from data obtained by their use as to the fertility of soils at present or in the near future. One of these methods is also frequently used for classifying soils with reference to their probable endurance. Results obtained by all methods are perhaps applicable to a discussion of the relation between chemical composition and physical texture. Our data seem to lead to certain conclusions with reference to all of these points and thus further justify their publication.

The origin, physical composition, behavior under crop, periodic results of water extractions and much other data concerning the soils discussed in this article are described elsewhere (7) in great detail. We shall limit our descriptions here to the mechanical analyses, but use such designating numbers and expressions as will enable the reader to refer readily to the article cited.

The soils used were all selected with reference to the United States Bureau of Soils classification. They comprised 5 silty clay loam soils of a single series and 8 fine sandy loams of different series. Our objective in selecting these soils was to obtain one group of as nearly identical physical texture and origin

as possible and another group which should belong to an identical class but differ in that the individual representatives belong to different soil series. The presence of the single clay loam is due to an erroneous preliminary physical classification. It is so very similar in its chemical properties to the silty clay loams that for most purposes we have grouped it with these latter. All soils have been cropped and under continuous observation for three years. They are kept in tight containers 30 by 60 inches in area, 18 inches deep, and watered with distilled water.

TABLE A

Mechanical Analysis. (Averages of two analyses)

CLASS	SILTY CLAY LOAMS						FINE SANDY LOAM							
Series	Yolo						Hanford	Fresno	Kimball	Tejunga	Madera	Arnold	Unnamed	Standish
Soil No.	1	2	3	4	5	6*	7	8	9	10	11	12	13	14
	per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
Fine gravel (1–2 mm.)	0.00	0.00	0.00	0.00	0.00	0.00	0.04	0.00	0.016	0.00	0.00	0.00	0.04	0.00
Coarse sand (1–1.5 mm.)	0.15	0.60	0.60	2.09	1.84	0.746	2.82	1.95	9.986	1.57	2.77	4.99	7.77	3.09
Medium sand (0.5–0.25 mm.)	0.23	1.50	1.04	1.24	1.70	0.574	2.87	3.19	6.308	2.94	2.818	4.38	14.55	4.05
Fine sand (0.25–0.010 mm.)	1.69	8.00	3.53	5.47	9.61	1.786	15.60	21.22	21.730	16.62	20.945	23.27	13.49	24.54
Very fine sand (0.01–0.05 mm.)	32.62	31.26	26.28	30.32	32.73	21.775	53.67	50.63	43.67	38.76	39.34	46.24	15.70	38.94
Silt (0.05–0.005 mm.)	42.68	36.97	38.12	37.99	30.52	37.415	16.01	17.57	10.87	26.25	19.075	9.61	34.72	15.55
Clay (0.005–0 mm.)	24.17	22.47	28.47	25.08	24.72	39.380	9.77	5.56	8.19	13.37	14.62	9.44	12.95	13.87
Sum of percentages	101.54	100.80	98.68	102.19	101.12	101.676	100.58	100.12	100.77	99.51	99.568	97.93	99.22	100.04
Gravel removed by previous screening	0.11	0.09	0.39	6.15	3.38	0.11	3.64	0.99	6.41	0.59	2.12	1.57	1.15	2.21

* Clay loam.

TIME OF EXAMINATION OF SOILS

The complete analyses and those of the extracts obtained by hydrochloric and citric acid were made only once and at the commencement of our experiments. The results by water extractions, however, varied from day to day and were very low with soils under crop at the height of the growing season. It is evident that determinations at no single date could represent the potential fertility of a soil. The data on water extractions are therefore the averages of the results of periodic examinations of uncropped soils in the year 1916 when the soils had been in place for an entire calendar year and had had every opportunity to come into equilibrium with their local environment.

FORM OF REPORTING RESULTS

Analyses of the hydrochloric and citric acid extracts are usually reported in the conventional forms: potash (K_2O), lime (CaO), magnesia (MgO) and phosphoric acid (P_2O_5), etc. Complete analyses are sometimes reported as the elements and sometimes in the conventional forms. Water extractions are usually reported in terms of parts per million of the various ions. For purposes of comparison of the different methods it is deemed desirable to report all results in terms of the conventional forms. Results by the first three methods are expressed as percentages, but those from the water extractions will be expressed as parts per million, since the conversion of these to percentages seems to subserve no useful purpose.

TABULATION OF DATA

The analyses by the complete (fusion) method and those by the hydrochloric-acid-extraction method comprised determinations of all of the usual constituents. Those by the citric acid and water extractions were confined to the elements potassium, calcium, magnesium and phosphorus. Nitrogen (NO_3) determinations on the water extracts were made and will be included for certain purposes. In tables 1 and 1A we include all of the results of analysis by the complete and hydrochloric acid methods for purposes of record only. The discussion will be based on the figures for the more important constituents, and since the results of the citric acid and water extractions are limited to these constituents, the results by all methods are included in tables 2 and 2A.

CHEMICAL CRITERIA AND PHYSICAL TEXTURE

A basic principle in the use of chemical criteria for estimating soil fertility is that soils of the same physical texture shall be compared among themselves. Obviously no two soils are of absolutely identical physical texture and the strict application of the principle would nullify the value of any comparison. To justify the utility of such comparisons we must assume that the soils compared, if not identical in physical texture, are either so similar that they would permit of equal root dispersion or so "light" that resistance to root dispersion becomes an insignificant factor in crop production. Our five silty clay loam soils belong to an identical series and class according to the United States Bureau of Soils classification. They are therefore quite similar in physical texture, and because of the special care taken in their selection, are probably more nearly alike than an equal number of soils of the same kind taken in the course of a soil survey. If there is any validity in the physical classification, the chemical criteria should be strictly comparable among themselves. The fine sandy loams belong to an identical class, but are from different series of the United States Bureau of Soils classification. These soils, however, are of a kind in which resistance to root dispersion is unquestionably low. Their chemical characteristics are therefore logically comparable.

TABLE 1

Complete analyses of soils (fusion method)

SOIL NUMBER	SERIES	CLASS	SiO_2	K_2O	Na_2O	CaO	MgO	Mn_3O_4	Fe_2O_3	Al_2O_3	P_2O_5	SO_3	VOLATILE	TOTAL
			per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
1	Yolo	Silty clay loam	60.81	1.77	1.66	1.51	4.53	0.38	7.12	14.03	0.17	0.09	8.05	100.12
2	Yolo	Silty clay loam	61.73	1.70	1.88	1.43	3.69	0.60	7.12	14.77	0.19	0.10	6.97	100.18
3	Yolo	Silty clay loam	60.45	1.85	2.07	1.50	3.57	0.41	7.32	14.15	0.21	0.05	8.43	100.01
4	Yolo	Silty clay loam	65.10	1.58	2.24	1.46	2.79	0.37	5.60	13.93	0.19	0.06	6.42	99.74
5	Yolo	Silty clay loam	65.29	1.98	2.74	1.48	2.66	0.36	5.36	13.41	0.22	0.06	6.31	99.87
6*	Yolo	Silty clay loam	61.02	1.88	2.16	1.90	3.03	0.38	7.76	14.19	0.17	0.06	7.91	100.46
7	Hanford	Fine sandy loam	62.04	2.54	2.76	3.92	2.53	0.46	5.84	15.63	0.24	0.03	3.55	99.54
8	Fresno	Fine sandy loam	68.46	2.92	3.40	3.19	1.04	0.40	4.16	14.60	0.15	0.03	1.87	100.22
9	Kimball	Fine sandy loam	66.28	3.34	2.62	2.35	1.54	0.43	6.40	14.38	0.19	0.03	3.02	100.58
10	Tejunga	Fine sandy loam	57.69	2.60	3.14	4.17	2.61	0.56	6.80	16.73	0.30	0.08	5.24	99.92
11	Madera	Fine sandy loam	56.95	1.84	2.68	4.53	3.79	0.42	7.44	16.85		0.04	5.04	99.58
12	Arnold	Fine sandy loam	64.16	2.60	3.64	3.81	1.34	0.44	4.64	16.21	0.20	0.02	2.89	99.95
13	Unclassified	Fine sandy loam	94.23	0.08	0.10	0.06	0.12	0.07	0.40	1.34	0.02	0.04	3.22	99.68
14	Standish	Fine sandy loam	58.61	1.60	2.70	4.12	1.67	0.50	6.08	18.29	0.22	0.05	5.46	99.30

* Clay loam.

TABLE 1A

Analyses by hydrochloric-acid extraction

SOIL NUMBER	SERIES	CLASS	INSOLUBLE	K_2O	Na_2O	CaO	MgO	Mn_3O_4	Fe_2O_3	Al_2O_3	P_2O_5	SO_3	VOLATILE	TOTAL
			per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
1	Yolo	Silty clay loam	70.64	0.82	0.22	1.36	3.59	0.32	7.04	9.34	0.17	0.06	6.97	100.53
2	Yolo	Silty clay loam	71.96	0.89	0.19	1.30	3.01	0.35	6.92	9.43	0.19	0.05	6.21	100.50
3	Yolo	Silty clay loam	70.49	0.89	0.21	1.48	3.32	0.35	6.96	8.84	0.20	0.05	7.17	99.96
4	Yolo	Silty clay loam	75.91	1.01	0.14	1.44	2.56	0.25	5.24	8.47	0.17	0.06	5.42	100.67
5	Yolo	Silty clay loam	75.69	1.05	0.19	1.43	2.46	0.25	5.00	7.77	0.22	0.06	6.07	100.19
6*	Yolo	Silty clay loam	70.81	0.81	0.17	1.86	2.72	0.32	7.70	9.39	0.16	0.04	6.52	100.50
7	Hanford	Fine sandy loam	79.94	0.81	0.26	2.08	1.84	0.32	5.60	6.03	0.18	0.03	3.09	100.18
8	Fresno	Fine sandy loam	90.18	0.31	0.11	1.30	0.86	0.27	3.00	2.44	0.11	0.03	1.90	100.51
9	Kimball	Fine sandy loam	83.10	0.67	0.17	1.71	1.03	0.22	6.32	4.67	0.19	0.03	2.29	100.40
10	Tejunga	Fine sandy loam	74.54	0.95	0.22	3.05	2.47	0.42	6.48	7.32	0.30	0.04	4.04	99.83
11	Madera	Fine sandy loam	75.86	0.80	0.15	2.23	2.60	0.17	6.08	8.05	0.18	0.04	3.59	99.75
12	Arnold	Fine sandy loam	85.66	0.49	0.13	0.56	0.96	0.07	4.08	5.78	0.20	0.02	2.00	99.97
13	Unclassified	Fine sandy loam	96.23	0.07	0.02	0.02	0.05	0.04	0.28	0.89	0.02	0.04	2.10	99.76
14	Standish	Fine sandy loam	75.28	0.74	0.18	2.44	1.07	0.41	5.12	9.92	0.11	0.05	4.27	99.38

* Clay loam.

TABLE 2

Silty clay loam soils of the same class and series. Arranged in order of crop (barley) producing power*

SOIL NUMBER	PRODUCTIVITY		COMPLETE ANALYSES				ACID EXTRACTIONS (HCl 1.115)				CITRIC ACID EXTRACTIONS (1 per cent)				SEASONAL AVERAGES OF H_2O EXTRACTIONS				
	General	Bushels per acre	K_2O	CaO	MgO	P_2O_5	K_2O	CaO	MgO	P_2O_5	K_2O	CaO	MgO	P_2O_5	K_2O	CaO	MgO	P_2O_5	NO_3.
			per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*p.p.m.*	†*p.p.m.*	*p.p.m.*	*p.p.m.*	*p.p.m.*
5	Good	88.4	1.98	1.48	2.66	0.23	1.05	1.43	2.46	0.22	0.039	0.452	0.220	0.101	57	127	40	12	146
1	Good	86.5	1.77	1.51	4.53	0.17	0.82	1.36	3.49	0.17	0.042	0.325	0.325	0.055	48	48	38	5	131
6*	Good	85.9	1.88	1.90	3.03	0.17	0.81	1.86	2.72	0.16	0.039	0.364	0.263	0.047	40	66	35	5	141
2	Good	84.0	1.70	1.43	3.69	0.19	0.89	1.30	3.01	0.19	0.041	0.336	0.164	0.069	66	52	25	3	120
4	Medium	70.4	1.58	1.46	2.79	0.19	1.01	1.44	2.56	0.17	0.034	0.450	0.224	0.051	54	118	43	7	141
3	Poor	56.2	1.85	1.50	3.57	0.21	0.89	1.48	3.32	0.20	0.039	0.422	0.144	0.072	52	45	23	5	88
Average composition			1.78	1.48	3.45	0.20	0.93	1.40	2.97	0.19	0.039	0.397	0.215	0.070	55	78	34	6	125
Variation (per cent)			6	2	16	8	8	4	12	8	4	14	22	20	8	46	24	40	14
Maximum individual variation (per cent)			11	3	31	15	*13	7	18	16	13	18	51	44	20	63	32	100	29

* Soil 6 (clay loam) is not included in the computations of averages and variations. It is placed in the table because of its general similarity, both physically and chemically, to the silty clay loams.

† p.p.m.=parts per million.

TABLE 2A

Fine sandy loam soils of the same class but different series. Arranged in order of crop (barley) producing power

SOIL NUMBER	PRODUCTIVITY		COMPLETE ANALYSES				ACID EXTRACTIONS (HCl 1.115)				CITRIC ACID EXTRACTIONS (1 per cent)				SEASONAL AVERAGES OF H_2O EXTRACTIONS†				
	General	Bushels per acre	K_2O	CaO	MgO	P_2O_5	K_2O	CaO	MgO	P_2O_5	K_2O	CaO	MgO	P_2O_5	K_2O	CaO	MgO	P_2O_5	NO_3
			per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*	*p.p.m.*	‡ *p.p.m.*	*p.p.m.*	*p.p.m.*	*p.p.m.*
8	Good	87.0	2.92	3.19	1.04	0.15	0.31	1.30	0.86	0.11	0.036	0.155	0.045	0.078	59	92	25	11	180
14	Good	80.8	1.60	4.12	1.67	0.22	0.74	2.44	1.07	0.11	0.036	0.264	0.104	0.040	63	71	28	9	96
11	Good	80.1	1.84	4.53	3.79	0.18	0.80	2.23	2.60	0.18	0.034	0.231	0.089	0.072	81	71	22	20	154
10	Medium	70.6	2.60	4.17	2.61	0.30	0.95	3.05	2.47	0.30	0.019	0.411	0.041	0.086	59	73	18	7	130
7	Medium	69.4	2.54	3.92	2.53	0.24	0.81	2.08	1.84	0.18	0.029	0.273	0.054	0.084	37	85	22	5	118
12	Poor	50.0	2.60	3.81	1.34	0.20	0.49	0.56	0.96	0.20	0.031	0.203	0.055	0.095	36	57	15	9	83
9	Poor	45.8	3.34	2.35	1.54	0.19	0.67	1.71	1.03	0.19	0.034	0.255	0.040	0.079	23	62	15	4	54
13*	Negligible		0.08	0.06	0.12	0.02	0.07	0.02	0.05	0.02	0.012	0.011	0.018	0.003	25	52	126	2	0
Average composition			2.49	3.73	2.07	0.21	0.68	1.91	1.55	0.18	0.031	0.256	0.061	0.076	51	73	21	9	116
Variation (per cent)			12	14	38	18	24	32	42	22	14	20	32	16	32	12	20	38	28
Maximum individual variation (per cent)			36	37	83	43	54	70	68	67	39	61	70	47	60	26	33	122	55

* Soil 13 is not included in computations of averages and variations. Its physical texture justifies classification as a fine sandy loam, but its obvious peculiarities and negligible yield make it undesirable to consider it in a determination of the characteristics of the class.

† In Soil 13 the water extractions constitute a single set of determinations.

Determination of the seasonal averages was unnecessary because of the low range observed and the lack of success in growing a crop

‡ p.p.m. = parts per million.

CHEMICAL ANALYSIS AND PRESENT PRODUCTION

If crop growth is dependent upon the ability of the soil to maintain the concentration of individual ions in the soil solution (1, 2), it would seem that any rational method should tend to show lower figures in soils of low production than in the better soils. The number of instances in which the poorer of a group of soils give lower results by any method than those from the better soils may be used as a rough measure of the rationality of the method. This, of course, is strictly true only in case all of the important soil constituents are included. None of the methods used, with the exception of the water extraction method, takes cognizance of the nitrate content of the soil. This being the case, the other methods must justify their use in soil fertility studies on the basis of the remaining constituents.

We have observed in our studies of periodic water extractions that a rise in nitrate content of a given soil is invariably attended by a rise of basic constituents and notably calcium, pointing to a close relationship between biological activities and the solutes found. In water extractions, the non-nitrate mineral constituents thus become of increased importance as an index of biological activity. No such advantage accrues to the other methods since the intensity of action of the reagents doubtless obscures the effects of organisms.

In tables 2 and 2a we have indicated by bold-faced figures those instances in which a given constituent of the poor or medium soils is lower than the same constituent in all of the good soils of the same group. The apparent deficiencies of both groups may be combined and summarized[1] in two ways.

First: Instances in which the poor and medium soils show lower figures than all of the good soils of their respective groups:

	K_2O	CaO	MgO	P_2O_5	TOTAL (EXCLUDING NO_3)	NO_3	TOTAL (INCLUDING NO_3)
Complete	1	1	0	0	2		2
Hydrochloric extraction	0	1	0	0	1		1
Citric extraction	4	0	3	0	7		7
Water extraction	3	3	4	3	13	3	16
Possible maximum	6	6	6	6	24	6	30

Second: Instances in which the poor soils show lower figures than all of the good soils of their respective groups:

	K_2O	CaO	MgO	P_2O_5	TOTAL (EXCLUDING NO_3)	NO_3	TOTAL (INCLUDING NO_3)
Complete	0	1	0	0	1		1
Hydrochloric extraction	0	1	0	0	1		1
Citric extraction	1	0	2	0	3		3
Water extraction	2	3	3	1	9	3	12
Possible maximum	3	3	3	3	12	3	15

[1] Soils 6 and 13 are not included in the summaries, although their inclusion would not change the conclusions fairly deducible therefrom.

The persistence with which low figures appear in water extractions of poor soils cannot be regarded as without significance. The same is true to a lesser degree of the citric-acid extractions, but certainly little can be said in favor of the use of either of the other methods for appraising the crop-producing power of soils.

We have purposely eliminated soil 13 from these comparisons for the reason that each and every constituent by the first three methods is very much below the same constituent in even the poorest of the other soils. All of the methods show serious deficiencies of the more important constituents so that any method would point out the limitations of this soil with reference to crop production. Indeed, a determination of silica alone should be indicative of its low fertility.

The use of chemical methods which are incapable of bringing out substantial differences between soils producing at the rate of 45 to 56 bushels of barley per acre and those producing at the rate of 80 or more bushels can hardly be justified. Water extraction does bring out substantial differences between soils; differences, moreover, which are less perceptible by other methods and which furthermore seem to be in general accord with the soils' crop-producing power.

It may perhaps be urged that a study which is limited to two classes of soil cannot have very general application. If, however, the chemical characteristics of the mineral components of soils have any influence on crop production, their effects should appear in each and every soil where they are not masked by adverse physical conditions of such magnitude as to constitute a limiting factor in crop production. Where such a condition exists no chemical method is adequate.

We are well aware that the consensus of opinion among soil investigators does not countenance the use of the complete analysis of the hydrochloric-acid extraction for predicting immediate crop yields. That they are being extensively used, however, and their application to this very purpose attempted, cannot be denied. We have ventured to present our data in the hope of still further limiting their use for such purposes.

COMPLETE ANALYSIS AND CONTINUED PRODUCTION

The complete chemical analysis has been extensively advocated as a means of forecasting the continued productivity of soils. Such a test is clearly adequate in the differentiation of soils like no. 13. All experience indicates that such soils will neither produce good crops at present without extraordinary additions of fertilizers or amendments, nor be capable of long-sustained productivity. Can we, however, justify the use of the complete analysis for

classifying soils which are sufficiently well supplied with the various important constituents to yield some crop, but nevertheless fall far below average productivity in total yield? Our soils with the exception of no. 13 represent such varying but not extreme degrees of fertility.

The complete analyses show that the poorer soils, no. 3, 9 and 12, carry totals of the same order of magnitude as the other soils of their respective groups and in spite of their comparatively low present production would be rated on an equality with the good soils by the complete analysis. All of these soils could doubtless have been brought to a high state of production by the incorporation of sufficient decomposable organic matter.[2] If the well recognized potency of organic matter in similar cases pertains to our soils, it is clear that both the present and future productivity of our poor soils is limited by the absence of an adequate nitrogen supply; or by the fact that the constituent minerals are of such a character that they will not replenish the soil solution at a sufficiently rapid rate unless they are broken down by indirect agencies such as those induced by decomposing organic matter. In either case, the limiting factor is not the totals shown by complete analysis, but the lack of absorbable forms of nitrogen or the resistant character of the constituent minerals. We take it that neither the present generation of farmers nor posterity are interested in speculations as to the relative endurance of soils which are not backed up by concrete evidence in terms of productiveness.

CHEMICAL COMPOSITION AND PHYSICAL CLASSIFICATION

Soils have from time immemorial been classified on the basis of their physical properties into such groups as clays, loams and sands. Classifications of this order have unquestionably justified their use, and, moreover, are probably in general accord with chemical composition when the soils so segregated are of the same climatic origin and environment. A question of very considerable interest is whether or not a more minute physical classification accords with differences in the chemical composition of the various groups. A comprehensive study of this question should obviously include a very large number of soils for determination of the probable variation of the various classes and individual soils within each class (5, 6). The large amount (3600 lbs.) of each soil requisite for our plot experiments necessarily limited the number of soils here reported upon. Our results, however, point to certain limitations on physical classification which may well be of extended application.

From tables 2 and 2A we have computed several sets of values, the significance of which may not be clear by inspection. "Variation" is the arithmetical mean of the individual percentage variations from the average of the group. The purpose of this figure is simply to obtain a convenient expression for measuring the extent to which the soils approach uniformity in composition. Computations of the mean probable variation by the method of least squares is of

[2]The integrity of our soils for other purposes has prevented a demonstration of this effect.

course not applicable to such a small number of soils. "Maximum individual variation" is, for each constituent, the percentage variation of that soil (highest or lowest) which deviates most from the average of all soils. Its purpose is to give an expression for measuring the extent to which individual soils deviate from the average for the group.

Soils of the same type (identical series and class)

The complete analysis may first be used as a test of uniformity in soils of the same or similar physical textures. We may then proceed to apply the figures obtained by the other methods (table 2).

The variation of the silty clay loams as shown by complete analysis is 16 per cent or less for all constituents. The maximum individual variations are of course higher. The hydrochloric-acid extraction gives results of the same order as the complete analysis as follows logically enough from the fact that the acid removes such a large proportion of the total of each ingredient. When we examine the results by the less rigorous extraction methods, differences of considerable magnitude occur. We believe that we have shown that these differences mean something in terms of crop production but they are not brought out by the physical classification. Opinion will perhaps differ as to the significance of the variations shown by complete and hydrochloric-acid extraction. It is clear, on the other hand, in view of the differences in yields of some of the soils and the great variations in the water extracts, that neither the crop-producing power of the soils nor their fertilizer needs could have been predicted from the physical classification alone.

Silty clay loam soils comprise a class of soils which represent a fairly advanced stage in the physical degradation of mineral particles. The same forces which tend to cause such changes in the size of the mineral particles are active in bringing together many different kinds of minerals. Fine soils in general are doubtless very heterogeneous in the origin of their constituent minerals, but since these consist largely of the complex silicates containing the same kinds of chemical elements, heterogeneous origin should lead to uniformity of ultimate chemical composition. Coarser soils derived from sedimentation processes might be as heterogeneous in origin and just as uniform in composition. Coarser soils "in place" would in general be derived from a more limited number of minerals and such soils from different rock formations would be likely to show greater variations in chemical composition than do the finer soils. There seems then good theoretical justification for assuming that the magnitudes of variation in the silty clay loams are at least no larger than those which would appear in the examination of other classes from the same or a different series, with the possible exception of soils of very fine texture, such as clays, which might be more uniform.

The variations found by us might have been materially lowered had we examined a very large number of soils. We do not mean to suggest that our

figures represent the probable mean variation. The individual variations, however, might and probably would have been even greater if more soils had been used.

Soils of different types (identical class but different series)

The figures (table 2A) for mean and maximum variation by complete analysis, hydrochloric and citric extraction are (with the single exception of P_2O_5 in the citric extraction) invariably higher than those shown by the Yolo silty clay loams. These differences are of considerable magnitude for most of the determinations. Apparently, then, there are greater differences, chemically as well as physically, between soils from different series of this class than between individual soils of the Yolo series of silty clay loams. To this extent the physical classification seems to be consistent, but since we have here only a single representative of each type we do not presume to offer a definite opinion on this point, nor can we extend such an opinion to terms of a generalization covering other types.

Soils of different classes

Here, again, our data are too meagre to serve as a basis for generalization. Comparison of the averages by complete analysis and hydrochloric-acid extraction shows substantial chemical differences (except in phosphoric acid) between two physical classes as here represented. It is perhaps not too much to assume that differences in ultimate chemical composition are likely to appear in comparisons of other classes. We hold, however, that chemical differences shown by complete analysis and hydrochloric acid do not determine either fertilizer needs or crop production. When we test the two classes by the figures for water extraction we see at once that the figures for a given constituent in individual soils of either class may run higher or lower than for the same constituent in individual soils of the other class. If we furthermore bear in mind that both classes include soils of good, medium and low production it is fairly obvious that physical classification into classes, as in the case of individual soils of an identical series and class, leaves much to be desired in the prediction of yields and prescription of fertilizers, however valuable it may be for determining general methods of cultivation.

CONCLUSIONS

The data presented heretofore indicate very clearly that the seasonal averages by water extraction of uncropped soils give figures more in accord with the soils' present crop-producing power than any of the usual methods. The large number of instances in which the soils of low production give low figures as compared with those obtained from soils of higher production is very striking. It is especially noteworthy that such extractions show substantial

chemical differences between soils which, on the average, vary between 50 and 80 bushels in yield. The method, therefore, is not limited to differentiation between soils of very high production and those which are totally unproductive, but may be applied to the great group of soils in which the farming population is most interested. We refer to soils so productive that the farmer cannot afford to allow them to remain uncropped, but not sufficiently productive to meet reasonable demands without fertilizers or amendments. We do not suggest that water extraction in its present development is of wide application to soils "in the field." One reason is that field soils are highly heterogeneous physically, chemically and in bacteriological flora and it is a matter of great difficulty to obtain a representative sample (8); another, that the condition of the soil at no single date is likely to represent its average condition during the growing season or at the period when temporarily depleted by plant withdrawals. W. H. Jordan in a recent paper (4) states:

> . . . . . that the treatment with water gives results more significant in their relation to the productiveness of the several soils than is the case with either of the acid solutions but even with the water, if there is any significance in the results obtained, it is shown in the amounts of total soluble matter rather than in the proportions of potassium and phosphorus compounds brought into solution. Not much can be claimed, however, for a method that shows as high solubilities for a soil producing 14.1 gm. of dry matter to the box as for a soil producing 96.8 gm. of dry matter per box.

We regard Dr. Jordan's statement as generally corroborative of our conclusions. The limitation which he suggests, however, cannot vitiate our more extended conclusions, since his extractions were made on some one given date (not stated); whereas the results obtained in this laboratory are averages from results on samples taken from the plots periodically throughout the year. Since the soil comprises a dynamic system and the extracts vary in magnitude from day to day (7), its potentialities are not to be appraised on a single set of determinations.

A corollary to our conclusions as to the superiority of the water-extraction method is that all of the other methods are entirely inadequate as measures of the present crop-producing power of soils. When we come to consider criteria of soil endurance we conclude that any reagent (fusion with alkalies, strong acids, etc.) which actively attacks the soil minerals will bring out substantial differences between highly silicious soils and those containing considerable percentages of bases; but will not sufficiently distinguish between good soils and those which, while somewhat productive, will not produce average crops without additions of fertilizers or amendments. No chemical analysis is either necessary or desirable to bring out the differences between soils which approach the extremes of mineralogical composition.

The number of soil classes considered here is too limited to permit of extended generalizations as to the correlation of chemical composition of soils with their physical textures. Even if we could have shown an absolute cor-

relation between ultimate chemical composition and physical texture (6), the great variation in yields and water extracts between individual soils within a type which is probably unusually uniform, and the further fact that soils of other types belonging to a very different class frequently give figures which fall within the limits of individual variation within a type, would cast grave doubts on the adequacy of physical classification for predicting probable yields or prescribing fertilizers. It is difficult to conceive that physical classification can be applicable to any other purpose than the determination of general methods of mechanical treatment.

To be of any great value to the farmer, the physical classification must at least be supplemented by extended data as to crop history and preferably by the application of chemical and biological criteria to each individual soil. These latter must be the most important function in any adequate investigation or survey with the possible exception of those cases in which the soils examined represent great extremes of chemical composition and physical texture.

ACKNOWLEDGMENTS

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The data from which the seasonal averages of the water extractions are computed were obtained by various members of the staff of this laboratory in the course of our seasonal studies. Thanks are extended to all concerned.

SUMMARY

1. Fourteen soils were studied with reference to chemical and mechanical composition and crop production under controlled conditions.

2. The soils included five individuals of a single series of one class and eight soils representing different series of another class.

3. The chemical treatment comprised complete analysis, hydrochloric-acid, citric-acid and water extractions.

4. Low figures by water extraction and to a lesser extent by citric-acid extraction are shown to be in general accord with the soils' crop-producing power.

5. Except in the case of highly silicious soils, the complete analysis and hydrochloric extractions are held to be worthless as criteria of present production or probable endurance.

6. Individual soils of a given series and class show less variation in chemical composition than do the representatives of different series of another physical class.

7. The great variation in crop yield and in the figures for water extractions, within a given series and class (soil type) indicates that physical classification into types is inadequate as a means of predicting probable yields or determining fertilizer requirements.

REFERENCES

(1) BURD, JOHN S. 1918 Water extractions of soils as criteria of their crop-producing power. *In* Jour. Agr. Res., v. 12, no. 6, p. 297–309.

(2) HALL, A. D., BRENCHLEY, WINIFRED E., AND UNDERWOOD, LILIAN M. 1913 The soil solution and mineral constituents of the soil. *In* Phil. Trans. Roy. Soc. London, ser. B, v. 204, no. 307, p. 179–200.

(3) HOAGLAND, D. R. 1918 The freezing-point method as an index of variations in the soil solution due to season and crop growth. *In* Jour. Agr. Res., v. 12, no. 6 p. 369–395.

(4) JORDAN, W. H. 1916 Measurements of soil fertility. N. Y. State Agr. Exp. Sta. Bul. 424, p. 389–412.

(5) PENDLETON, R. L. 1918 Are soils mapped under a given type name by the Bureau of Soils method closely similar to one another? *In* Univ. Cal. Pub. Agr. Sci., v. 3, no. 11.

(6) ROBINSON, W. O., STEINKOENIG, L. A., AND FRY, W. H. 1917 Variation in the chemical composition of soils. U. S. Dept. Agr. Bul. 551.

(7) STEWART, G. R. 1918 Effect of season and crop growth in modifying the soil extract. *In* Jour. Agr. Res, v. 12, no. 6, p. 311–368.

(8) WAYNICK, D. D. 1918 Variability in soils and its significance to past and future soil Investigation. *In* Univ. Cal. Pub. Agr. Sci., v. 3, no. 8, p. 135-162.

THE INFLUENCE OF AMMONIUM SULFATE ON THE GERMINATION AND THE GROWTH OF BARLEY IN SAND AND SOIL CULTURES KEPT AT DIFFERENT MOISTURE CONTENTS AND AT VARIOUS OSMOTIC CONCENTRATIONS OF THE SOIL SOLUTION[1]

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INTRODUCTION

An adequate supply of nitrogen in the soil is one of the most important questions in soil fertility. This is true not only as a theoretical consideration, nitrogen being one of necessary elements for plant growth, but also from the practical point of view, because nitrogen in normal times is the most expensive of the fertilizing materials.

The inorganic nitrogenous materials which are available for agricultural purposes are very limited in both variety and amount. We have nitrate of soda, the natural supply of which will necessarily be declining in the course of time; sulfate of ammonia, the manufacture of which as a by-product in the coal industries is rapidly expanding; and calcium nitrate and calcium cyanamid, as the products of fixation of atmospheric nitrogen with the aid of electrical energy.

There undoubtedly exist a set of conditions under which a given fertilizer acts at its best. It is believed that this optimum set of conditions is specific for each fertilizer, and that it may or may not be the same for different nitrogenous materials. It is of prime importance, therefore, that one should know the behavior of that fertilizer under different conditions that go to modify its action, so as to make it most effective in crop production.

The present study is made of ammonium sulfate in order to throw some light on its fertilizing value under some variable conditions as they may be found in agricultural practice.

REVIEW OF LITERATURE

Ammonium sulfate is one of the few salts regarding whose fertilizing value there exist so many divergent opinions. Since the middle of the last century, when Boussingault (26), Ville (231), and Lawes, Gilbert and Pugh (128) performed their celebrated experiments on the sources of nitrogen of vegetation, and up to the present time, the question remains in dispute whether ammoniacal nitrogen is equal or inferior in its efficiency to nitrate nitrogen. Establish-

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ing the fact that nitrate nitrogen can be utilized without first being converted into the ammonia form, Ville (231) has recorded at the same time that with potassium nitrate for the nitrogen supply the plants contained more nitrogen than those with the sal ammoniac (ammonium chloride). From this he concluded that for plant growth ammoniacal nitrogen was inferior to nitrate nitrogen.

With the progress of agricultural research there had accumulated a vast amount of literature recording the data which throw some light on the question of efficiency of ammoniacal nitrogen versus nitrate nitrogen for the growth and development of agricultural plants.

Lawes and Gilbert (129) were among the first to investigate the relative value of ammonium sulfate and sodium nitrate for the growth of field crops. Investigations with wheat and barley were started by them as early as 1852, followed by oats in 1876 and continued up the present time. Each crop was grown on the same plot year after year. The results, on the whole, show that for plant growth nitrate of soda is preferable to the ammoniacal nitrogen, which was supplied with a mixture of the equal amounts of ammonium sulfate and ammonium chloride.

In 1880 Wagner (248) proposed a pot method for studying the efficiency of different nitrogenous fertilizers and manures. He and his co-workers (249), 250, 251), over a period of many years studied these fertilizers on many soils in Germany with different crops. His general conclusion was that, if nitrate of soda nitrogen is given a value of 100, the ammonium sulfate nitrogen was equal to 90, taking all the crops and soils into consideration.

In this country the investigations in Pennsylvania (103, 104) are among the oldest on this subject. These experiments have been carried on since 1880 on field plots, on which a general field rotation is maintained. In the first ten years of the experiment (104) ammonium sulfate gave better results than nitrate of soda, but as an average for over thirty years (104), the nitrate of soda was superior to ammonium sulfate.

Similar results were obtained by the Massachusetts Agricultural Experiment Station. The data of 25 years (66) show that, under the conditions of the experiment, ammonium sulfate was only 88 per cent as efficient as nitrate of soda.

New Jersey's (135) 15-years results with all the crops in a 5-year rotation show that the efficiency of ammonium sulfate was equal to 84.6 in connection with minerals alone and 91.6 in connection with solid manure, leached, if a value of 100 is assigned to the nitrate of soda.

The investigations of the Rothamsted and the Woburn (238) stations in England, of Wagner and his co-workers in Germany, and of the New Jersey, Pennsylvania and Massachusetts stations in this country, are perhaps, among the most authoritative on the subject of efficiency of these two fertilizing materials. There are a host of others, who found the superiority in the value of nitrate of soda over that of ammonium sulfate for the nitrogenous fertilizer

and whose results are no less trustworthy than those that have been mentioned above.

Among these investigators can be mentioned Sachs and Knop (187), Towar (227), Grahl (75), Warington (254), Kirchner (118), Voorhees (242, 243, 244), Voorhees and Lipman (245, 246), Lipman and his associates (133, 134, 135, 136, 137 and 138), Grandeau (76, 77), Bolin (23, 24), Kretschmer (126), Feruglio (61), Grazia and Galdieri (79), Schneidewind (190, 191, 192, 193, 194, 195, 196), Ward (253), Cillis (42), Porter and Gaut (170), Chodat (41), Schulze (198), Schmoeger (189), Steiglich (214), Söderbaum (209, 212), Stutzer (216, 217), Schreiber (197), Bachmann (15), Crowther (49, 50), Gilchrist (70), Berry (20), Vanha (229), Rhodin (182), Bremer (32) (for top-dressing), Buchner (37), Svoboda (219), Hansen (84), Malpeaux (140, 141), Rindell (178), de Jong (110, 111), Patterson (165), Larsen (127), Parsons (164), Woods (269), Wheeler and Adams (261), Hunt and his co-workers (103, 104), Goessman (73), Brooks and his associates (33, 34), Tacke (222), Duggar and Caethen (55), Kelkar (114), Ballard and Volck (17), Gerlach (69), Bonomi (25), Voelcker (237, 240), Herke (98), (Kappen 113), Bauman (18), Niklevski (156), McClelland (148), Popp (169) and many others.

On the other hand, there are just as many men who have found ammonium sulfate to be either equal or superior to nitrate of soda in plant production. Among these investigators can be mentioned Hellriegel (94), Kloepfer (120, 121, 122), Kraus (123, 124, 125), Clausen (44, 45, 46), Rhodin (180, 181, 182, 183), Huston (106), Vikier (232), Bovell and D'Albuquerque (29), Kelkar (114), Bachman (11, 12, 13, 16), Otto (160, 161), Lilienthal (130, 131), Caruso (39), Voelcker (233, 235, 239), Kretschmer (126), Wein (260), Watts (256, 257), Sebelien (201), Weibull (259), Süchting (218), Bolin (24), Kleberger (119), Wagner (252) (on fire-holding power in tobacco), Söderbaum (210, 211), Buchner (37), Hendrick (97), Swanwick and Kinch (220, 221), Jackometti (108), Harper (85), Milburn (150), Nazari (155), Blobel (22), Bauwens (19), Harrison (88, 89), and others (90), Rawson (177), Gilchrist (71, 72), Greig (80), Mayer (146) D'Albuquerque and Bovel (51), Bovell (30), Wilcox, Kelley and Krauss (265), Orchinnikov (158), Damseaux (52), Malpeaux (141), Tempany (224), Stokes (215), Etheridge (57), Wright (270), Kelley (115, 116), Anstead (10), Gardner and Brown (64), Van Hoek and Ranwerda (230), Calvin (38), Milburn and Gaut (151), Gaul (67, 68), Kimbrough (117), Wehnert (258), Rindell (178), Mausberg (143, 144), Haselhoff (91), Easterby (56), Oswald and his associates (159), Hiltner and Lang (101), Lipman and his associates (137), von Feilitzen (60), Lipman and Gericke (130), Porter and McWilliam (171), Schmitz (188), and many others (1, 2, 3, 4, 5, 6, 7, 8, 9).

Comparing the names of these groups we find some of them to appear in both. In many different instances the results of the same men are sometimes in favor of nitrate of soda and sometimes in favor of sulfate of ammonia. In some cases the differences are insignificant, while in others they are very considerable. The inferiority of ammonium sulfate in the Rothamsted and the

Woburn experiments in England, and at the New Jersey, Pennsylvania, and Massachusetts stations in this country, are the most noteworthy. In every case of the field experiments the yield of crops grown with ammonium sulfate as the source of nitrogen was fair at the beginning. At the Pennsylvania Station where the soil contained a fair amount of lime, the yields for 10 years or more on the ammonium sulfate plots were larger than those on the nitrate of soda plots. The yields on the ammonium sulfate plots, however, declined in time. This naturally led to investigations as to what was the cause of this gradual decline in the efficiency of the ammonium sulfate. In nearly every case it is now attributed to a deficiency of lime in these soils, for when lime was applied to these soils either in the field or in pot tests there was a considerable increase in the crop yield. In this connection reference may be made to Voelcker (234, 241), who studied the soils at Woburn; Hall (81) at Rothamsted; Brown (35), Brown and his associates (36), White (262), and Gardner and his associates (65), in Pennsylvania; and Blair and McLean (21) in New Jersey. Ruprecht and Morse (184) studying the soils from Massachusetts experimental plats came to the conclusion that the ill effect of ammonium sulfate was due to the formation of sulfates of iron and alumium, which takes place in the absence of the available compounds of calcium.

It is curious enough that in several of these experiments no provision was made to replenish the lime in the soil, although other fertilizers were applied year after year. Since ammonium sulfate, from the very nature of the salt, has the tendency to increase the soil acidity, it caused the accumulation of acids in the soils to such an extent that normal crop growth was almost impossible. When some lime was added, however, as was the case in the Pennsylvania (262, 65) experiments, the yields of crops of the ammonium sulfate plots came nearly up to the normal.

Somewhat similar conditions affected Wagner's results (251). He worked with a great variety of soils, some of them very deficient in lime. Yet in experimenting with most of these soils, he, while applying both potassium and phosphorus, supplied no lime in any form. When lime was introduced in his pot experiments the results for ammonium sulfate improved considerably, sometimes exceeding those for sodium nitrate.

Experimenting with nitrate of soda, sulfate of ammonia, nitrate of lime and calcium cyanamid on Rothamsted soils, Hall (81) finds very little difference between these nitrogenous materials in so far as crop growth is concerned. The differences, if they exist, do not exceed 10 per cent of the total crop. Indeed, in most instances, when ammonium sulfate is compared with sodium nitrate the difference in their effectiveness for plant growth is very small. It is only in special cases, as pointed by Hall (82), as on very light sands, heavy clays and soils very short of lime, that secondary considerations do come in and modify the results of the experiment.

Again, it is universally recognized that nitrate of soda is a one-season fertilizer. For this reason in the 1-year experiments nitrate of soda and ammonium

sulfate are hardly comparable, since the latter has a considerable after-effect (113, 183, 15, 125, 93, 100, 255).

There is still another feature which may be mentioned in connection with the present discussion. Russell (186), speaks of the toxicity of ammonium sulfocyanide, which in former years was frequently an impurity in ammonium sulfate manufactured from gas liquor. When present in sufficient quantities it caused an injury to the plants. "It is rarely," says Russell (184), "if ever, found now." Just how much of some of the ill effect noticed in connection with the use of ammonium sulfate could be attributed to this cause is impossible to say at present. Yet, it could hardly be denied that the methods in the manufacture of this salt in the earlier days of the industry were more crude than they are now, and it is very possible, therefore, that some of the impurities did play some part in the effectiveness of this salt as a fertilizer.

Consideration may now be given to the form of nitrogen which can be best assimilated by plants. As early as 1867 Hampe (83) observed that the growth of plants with ammonium salts as the source of nitrogen was rather slow at first, but was accelerated later. A similar observation was reported by Wagner (247) a year later. Evidently, it was the starting point from which has gradually developed a notion that ammoniacal nitrogen could hardly be used as such. Since the ammonium salts are quickly converted into a nitrate form, it was later taken for granted that ammonium sulfate, for instance, could not be used as such. On this account, the efficiency of the nitrogen of this salt was considered to be impaired. If it were true, the argument would be a very sound one, for it takes some time for the nitrification to take place. Moreover, the change of the nitrogen from the ammonia to a nitrate form is hardly ever 100 per cent efficient.

The fallacy of the conception, however, is now fairly well established. Over 30 years ago Pitsch (168) demonstrated that with nitrification excluded oats can grow and normally develop with ammonium sulfate as its source of nitrogen. Since then a number of other investigators working with different plants and media have obtained similar results. For an extensive review of the literature on the subject reference may be made to an excellent article of Hutchison and Miller (107). Among the latest contributions, with further verification that higher plants can assimilate ammonia nitrogen directly, may be mentioned Pantanelli and Severini (162, 163), Petrov (167), Kalinkin (112), Periturin (166), Shulov (207), Ritman (179), Prianishnikov (174, 175, 176), and Morozov (154). It is true that some plants are found to be able to assimilate the ammonia nitrogen more readily than others. Nikolaeva (157), even reports that one of the lupines was unable at all to use ammonia nitrogen. Yet, in the majority of cases no difficulty was encountered in growing various higher plants without nitrates.

This fundamental question of the direct assimilation of ammonia nitrogen by agricultural plants is considerably modified in field practice by the fact previously mentioned that at least a portion of the ammonia nitrogen in most

of the common soils under normal cultivation is very quickly changed into nitrate nitrogen.

The conditions under which any fertilizer acts may modify the efficiency of that fertilizer to a considerable degree. The extreme variations in the effect of ammonium sulfate is often attributed to different conditions of the investigation.

Besides the reaction of the soil, already mentioned above, the time and the manner of the application, the character of the season, the moisture content of the soil, temperature, etc., are among the factors which may enhance or diminish the effectiveness of ammonium sulfate, as noticed by Bachmann (18). De Jong (109) has reported that the efficiency of ammonium sulfate was very high in very wet seasons, as compared with that of dry seasons. Pospisil (172), comparing ammonium sulfate with sodium nitrate during four years, two of which had dry and two wet growing seasons, found that in the dry seasons nitrate of soda gave better results than ammonium sulfate, and in the wet seasons ammonium sulfate was superior to sodium nitrate.

The relationship of moisture content of the soil to the yield of crop was studied by many investigators. Hellriegel (95) found that the best yield of barley grain in sand cultures was obtained at 40 per cent of water-holding capacity, while the best yield of straw was reached at 60 per cent. Wollny's (268) results show that the optimum moisture content of soils for several farm crops lies between 60 and 80 per cent of the water-holding capacity. Similar results are recorded by Mayer (145) who also found that in this respect there exist some variations for different agricultural crops. Von Seelhorst (202) has shown that the effectiveness of fertilizers increased with the increase in moisture content. Similar results were reported by Morgan (153) and Colebatch (47). Also de Grazia (78), Harris (87) and Pospisil (172) came to the same conclusion, namely, that the action of fertilizers is influenced by the moisture content.

Von Seelhorst, alone and with his associates (203, 204, 205), Prianishnikov (173), Hunter (105), Tulaikov (228), Widtsoe (263), Widtsoe and Merrill (264) also considerably contributed to our knowledge of the subject. The results of Mitscherlich (152), however, differ from those of other investigators in so far as they show that with potatoes, oats and peas grown in quartz sand, two soils and muck, the crop yield continued to increase with an increase of the water-holding capacity up to 100 per cent.

In view of the foregoing facts it seemed to be advisable to investigate more closely the relation of the moisture content of the soil to the effect of ammonium sulfate in the germination, growth and the development of some agricultural plant. How does a given agricultural plant, when subjected to different applications of the necessary, or even the unnecessary, elements that go to make plant tissue, react when the moisture supply is varied within the limits often met in normal field operations?

In order to throw some light on this question, which is important for its general theoretical interest, and also for the practical bearing on soil management, the present investigation was undertaken.

EFFECT OF MOISTURE CONTENT OF SEA SAND ON THE GROWTH OF BARLEY, WITH AMMONIUM SULFATE AS THE SOURCE OF NITROGEN

Procedure

In this work the chemically pure (Baker's analyzed) substances were used for the plant nutrients. The only exception was calcium carbonate, which was reprecipitated powder, U. S. P.

After some preliminary experiments the following procedure was adopted. The plants of barley were grown in pot cultures in sea sand as a medium. Four different series of pots were prepared, each consisting of 54 pots. The moisture content in the first series was kept at 20 per cent; in the second series, at 40 per cent; in the third series, at 60 per cent; and in the fourth series, at 80 per cent of the water-holding capacity. The water-holding capacity of the sand was 24.33 per cent (an average of three determinations), as determined by the funnel method. The pots were of glazed earthenware and were 25 cm. in height and 25 cm. in inside diameter. Two kilograms of sand was used in each pot. The hygroscopic moisture of the sea sand was only 0.05 per cent, and it was disregarded in calculating the amount of water to add to each pot. Table 1 gives the per cent and the total amount of water added to the pots of each series. To find the absolute per cent of water at which each series was kept it is only necessary to add the hygroscopic moisture (0.05 per cent) to the per cent given in the table.

TABLE 1

Moisture content in the sea sand cultures of different series

PER CENT OF WATER-HOLDING CAPACITY	PER CENT BASED ON DRY SAND	AMOUNT OF WATER PER POT
		cc.
20	4.86	97.2
40	9.72	194.4
60	14.58	291.6
80	19.44	388.8

The moisture content of the sea sand cultures was kept as nearly constant as possible by adding water every day. Every other day the pots were weighed and brought to the original moisture content. On the days when the pots were not weighed the water was added to each pot in approximate proportion to the water loss on the preceding day.

The sea sand contained practically no nutrient material, as will be seen from the growth of the plants in the control pots. The cultures, therefore, were treated with a complete nutrient solution. The treatment consisted of

ammonium sulfate, monopotassium phosphate, magnesium sulfate and ferrous sulfate. These ingredients were added in solution, while calcium carbonate was mixed with sand previously to the introduction of the solution. Besides the variations in the moisture content, the fertilizing materials were varied as follows: ammonium sulfate was added from 0.05 to 0.40 gm. per pot; calcium

TABLE 2

Application of salts per pot containing 2 kgm. of sea sand, and the calculated values in pounds per acre

POT NO.	$(NH_4)_2SO_4$		$CaCO_3$		KH_2PO_4		$MgSO_4$		$FeSO_4.7H_2O$		$NaNO_3$		$Ca(NO_3)_2$	
	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre
	gm.	*lbs.*	*gm.*	*lbs.*	*gm.*	*lbs.*	*gm.*	*lbs.*	*gm.*	*lbs.*	*gm.*	*lbs.*	*gm.*	*lbs.*
0														
1	0.05	50	0.50	500	0.40	400	0.20	200	0.05	50				
2	0.05	50	1.00	1000	0.40	400	0.20	200	0.05	50				
3	0.05	50	2.00	2000	0.40	400	0.20	200	0.05	50				
4	0.05	50	0.50	500	0.80	800	0.20	200	0.05	50				
5	0.05	50	1.00	1000	0.80	800	0.20	200	0.05	50				
6	0.05	50	2.00	2000	0.80	800	0.20	200	0.05	50				
7	0.10	100	0.50	500	0.40	400	0.20	200	0.50	50				
8	0.10	100	1.00	1000	0.40	400	0.20	200	0.05	50				
9	0.10	100	2.00	2000	0.40	400	0.20	200	0.05	50				
10	0.10	100	0.50	500	0.80	800	0.20	200	0.05	50				
11	0.10	100	1.00	1000	0.80	800	0.20	200	0.05	50				
12	0.10	100	2.00	2000	0.80	800	0.20	200	0.05	50				
13	0.20	200	0.50	500	0.40	400	0.20	200	0.05	50				
14	0.20	200	1.00	1000	0.40	400	0.20	200	0.50	50				
15	0.20	200	2.00	2000	0.40	400	0.20	200	0.50	50				
16	0.20	200	0.50	500	0.80	800	0.20	200	0.50	50				
17	0.20	200	1.00	1000	0.80	800	0.20	200	0.05	50				
18	0.20	200	2.00	2000	0.80	800	0.20	200	0.05	50				
19	0.40	400	0.50	500	0.40	400	0.20	200	0.05	50				
20	0.40	400	1.00	1000	0.40	400	0.20	200	0.05	50				
21	0.40	400	2.00	2000	0.40	400	0.20	200	0.05	50				
22	0.40	400	0.50	500	0.80	800	0.20	200	0.05	50				
23	0.40	400	1.00	1000	0.80	800	0.20	200	0.05	50				
24	0.40	400	2.00	2000	0.80	800	0.20	200	0.05	50				
L			3.00	3000	0.60	600	0.20	200	0.05	50	0.50	500		
S					0.60	600	1.00	1000	0.05	50			0.35	350

[1] L and S stand for Lipman's and Shive's cultures, respectively.

carbonate, from 0.05 gm. to 2.0 gm.; and monopotassium phosphate, from 0.4 gm. to 0.8 gm. per pot. The treatment of each pot is given in table 2, which also gives the values in pounds of different salts per acre, as based on 2,000,000 pounds of soil (approximate weight of an acre of soil 6 inches deep).

Besides the control pots, which were left untreated, two pots in each series designated as 20L, 40L, 60L, and 80L, respectively, were treated with a modified

TABLE 3

Dry weight of barley grown in sea sand cultures of different moisture contents, total water loss per pot, and amount of water required to produce 1 gm. of dry matter under different treatments of salts

POT NO.	DRY WEIGHT OF TOPS			TRANSPIRATION AND EVAPORATION			WATER REQUIREMENTS
	A	B	Average	A	B	Average	Average
Series of 20 per cent of water-holding capacity							
	gm.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*
200	0.0415	0.0680	0.0558	200	219	210	3,763
201	0.2620	0.2700	0.2660	290	278	284	1,068
202	0.4150	0.3450	0.3800	403	417	410	1,078
203	0.3520	0.4575	0.4048	426	466	446	1,101
204	0.1240	0.2545	0.1893	169	246	208	1,098
205	0.2380	0.2900	0.2640	251	293	272	1,030
206	0.2820	0.3170	0.2995	339	331	335	1,119
207	0.2910	0.3285	0.3098	310	381	345	1,116
208	0.4880	0.4130	0.5405	468	429	449	991
209	0.5120	0.4250	0.4685	497	410	454	950
210	0.3100	0.3250	0.3175	306	413	370	1,166
211	0.2838	0.3998	0.3418	322	395	359	1,050
212	0.3230	0.4910	0.4070	369	432	401	985
213	0.4200	0.3317	0.3759	474	433	454	1,207
214	0.4740	0.5580	0.5110	514	541	528	1,033
215	0.2610	0.3160	0.2885	359	319	339	1,174
216	0.4200	0.4740	0.4470	413	432	423	946
217	0.3832	0.4400	0.4116	367	384	376	913
218	0.4330	0.1913	0.3122	398	222	310	993
219	0.4295	0.2625	0.3460	445	310	378	1,092
220	0.4980	0.5410	0.5195	518	623	571	1,099
221	0.4670	0.1920	0.3295	362	179	271	823
222	0.1995		0.1995	189		189	947
223	0.4230	0.2190	0.3210	332	213	273	850
224	0.3640	0.4885	0.4263	300	351	326	765
20S	0.4130	0.4260	0.4195	461	468	465	1,181
20L	0.4725	0.5825	0.5275	493	682	588	1,114
Series of 40 per cent of water-holding capacity							
400	0.0710	0.0775	0.0743	543	522	533	7,174
401	0.2745	0.2545	0.2645	642	723	683	2,578
402	0.4260	0.3045	0.3653	674	685	680	1,861
403	0.3425	0.3325	0.3375	678	684	681	2,018
404	0.3015	0.2415	0.2715	667	546	607	2,236
405	0.3015	0.2865	0.2940	641	632	637	2,167
406	0.4745	0.2950	0.3848	705	622	664	1,725
407	0.4010	0.3615	0.3813	670	665	668	1,753
408	0.4730	0.4720	0.4715	721	779	750	1,591
409	0.5070	0.4540	0.4805	781	786	784	1,633
410	0.3620	0.2505	0.3060	634	590	612	2,000
411	0.4860	0.4985	0.4923	667	740	704	1,430
412	0.5565	0.5960	0.5763	873	748	811	1,406

TABLE 3—*Continued*

POT NO.	DRY WEIGHT OF TOPS			TRANSPIRATION AND EVAPORATION			WATER REQUIREMENTS
	A	B	Average	A	B	Average	Average
	gm.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*
413	0.7245	0.6405	0.6825	851	900	876	1,284
414	0.6585	0.6540	0.6563	873	852	865	1,320
415	0.6940	0.6860	0.6900	927	972	950	1,377
416	0.7655	0.6505	0.7080	760	768	764	1,079
417	0.6540	0.7135	0.6838	807	829	818	1,196
418	0.8415	0.6865	0.7640	1,038	905	972	1,272
419	0.4510	0.5840	0.5675	665	832	749	1,320
420	0.8530	0.7445	0.7988	1,092	1,042	1,067	1,335
421	0.4575	0.5570	0.5073	553	1,122	838	1,653
422	0.5318	0.6817	0.6068	696	674	685	1,128
423	1.1095	0.8130	0.9613	1,086	870	978	1,018
424	0.9665	0.9265	0.9465	1,165	1,017	1,091	1,152
40S	0.6455	0.6980	0.6718	823	893	858	1,277
40L	0.7316	0.8097	0.7707	1,112	1,117	1,115	1,446
Series of 60 per cent of water-holding capacity							
600	0.0600	0.0445	0.0523	828	713	771	14,742
601	0.1995	0.2010	0.2003	681	459	570	2,850
602	0.1490	0.2460	0.1975	504	621	573	2,901
603	0.2405	0.3040	0.2723	774	582	678	2,493
604	0.3150	0.2925	0.3038	874	642	758	2,493
605	0.4025	0.3410	0.3719	841	693	767	2,062
606	0.3455	0.4155	0.3805	831	851	841	2,210
607	0.3980	0.5660	0.4820	611	674	643	1,334
608	0.5755	0.6145	0.5950	730	858	794	1,320
609	0.6490	0.5060	0.5775	695	646	671	1,162
610	0.5405	0.4905	0.5155	678	555	617	1,197
611	0.5210	0.6000	0.5605	884	639	762	1,359
612	0.5432	0.7660	0.6546	870	833	852	1,321
613	1.0090	1.0380	1.0235	1,190	1,282	1,236	1,208
614	1.0115	1.2070	1.1093	1,233	1,329	1,281	1,155
615	1.1050	1.1980	1.1515	1,164	1,391	1,278	1,110
616	0.7430	0.8110	0.7770	1,088	965	1,027	1,644
617	0.9825	1.0700	1.0263	1,232	1,170	1,201	1,171
618	1.2555	0.9405	1.0980	1,294	1,143	1,219	1,111
619	1.1880	1.2350	1.2115	1,223	1,487	1,355	1,119
620	1.0870	0.8475	0.9673	1,376	1,293	1,335	1,385
621	1.2880	1.1260	1.2070	1,467	1,391	1,429	1,184
622	0.7095	1.2150	0.9623	992	1,168	1,080	1,123
623	1.1460	1.2220	1.1840	1,277	1,284	1,281	1,082
624	1.1090	1.1345	1.1218	1,505	1,498	1,501	1,338
60S	1.3695	1.4495	1.4095	1,347	1,420	1,384	982
60L	1.4877	1.5010	1.4944	1,893	2,245	2,068	1,383

TABLE 3—*Concluded*

POT NO.	DRY WEIGHT OF TOPS			TRANSPIRATION AND EVAPORATION			WATER REQUIREMENTS
	A	B	Average	A	B	Average	Average
	Series of 80 per cent water-holding capacity						
	gm.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*cm.*
800	0.0700	0.0755	0.0728	721	791	756	10,385
801	0.5140	0.2975	0.4058	927	820	864	2,128
802	0.4895	0.4300	0.4598	958	986	972	2,113
803	0.5890	0.5045	0.5468	1,081	1,018	1,050	1,920
804	0.4200	0.3075	0.3637	950	825	888	2,440
805	0.4195	0.4360	0.4278	907	892	900	2,103
806	0.4350	0.5260	0.4805	947	1,029	988	2,059
807	0.5005	0.4605	0.4805	942	1,030	986	2,054
808	0.5370	0.7430	0.6400	970	1,014	992	1,550
809	0.8715	0.8915	0.8815	1,090	1,250	1,170	1,327
810	0.5965	0.5960	0.5963	980	956	968	1,624
811	0.6265	0.6468	0.6382	1,009	979	994	1,558
812	0.5270	0.7900	0.6585	1,044	1,089	1,067	1,620
813	0.9620	0.8580	0.9100	1,241	1,300	1,271	1,397
814	1.0820	1.0500	1.0660	1,358	1,511	1,435	1,346
815	1.1183	1.3403	1.2293	1,455	1,496	1,476	1,202
816	0.9765	0.4110	0.6938	1,290	966	1,128	1,625
817	0.7030	1.0745	0.8888	1,218	1,327	1,273	1,432
818	1.1850	1.0755	1.1303	1,456	1,348	1,402	1,241
819	1.0350	0.5582	0.7966	1,413	1,151	1,282	1,609
820	1.1460	0.5675	0.8568	1,572	1,155	1,364	1,592
821	1.4210	1.2405	1.3308	1,776	1,847	1,812	1,361
822	1.4510	1.2060	1.3285	1,504	1,391	1,448	1,090
823	1.5320	1.2978	1.4199	1,702	1,508	1,605	1,130
824	1.9940	1.4690	1.7318	2,055	1,802	1,929	1,114
80S	1.6375	1.0075	1.3225	1,584	1,575	1,580	1,194
80L	1.8393	1.4415	1.6404	2,908	2,355	2,632	1,605

Lipman's (136) formula, consisting of 0.5 gm. sodium nitrate, 3.0 gm. calcium carbonate, 0.6 gm. monopotassium phosphate and 0.05 gm. ferrous sulfate. For additional comparison, eight more cultures were treated with a nutrient solution, which approximately equals Shive's (206) culture R_5C_2, as added in a single application at 60 per cent of water-holding capacity. These cultures were designated as 20S, 40S,60S, and 80S, respectively.

The plants were germinated in moist sand and, when grown to about 2 inches in height, transplanted into pots. Six plants, selected for uniformity in transplanting, were grown in each pot. The plants were harvested after 30 days of growth in culture pots, dried in the oven for 2 days at 90°C. and then for 12 hours at 105°C. After cooling, the plants were weighed. The weights, together with data on transpired and evaporated water, and the water requirement of plants are given in table 3, and illustrated in figure 1.

Results and Discussions

On examining figure 1 and the values in table 3 one notices that there is a considerable variation in plant growth due to the different moisture contents under which the plants were grown. This is especially noticeable in culture pots from numbers 13 to 24 and including the cultures of Lipman (136) and of Shive (206). In some instances the variation between the culture pots kept at different percentages of moisture with the same fertilizer treatments is greater than the variations in the same series due to differences in applied fertilizer. In the majority of cases the yield from pots of the 20 per cent series is lower than that from the corresponding pots of the 40 per cent series. The

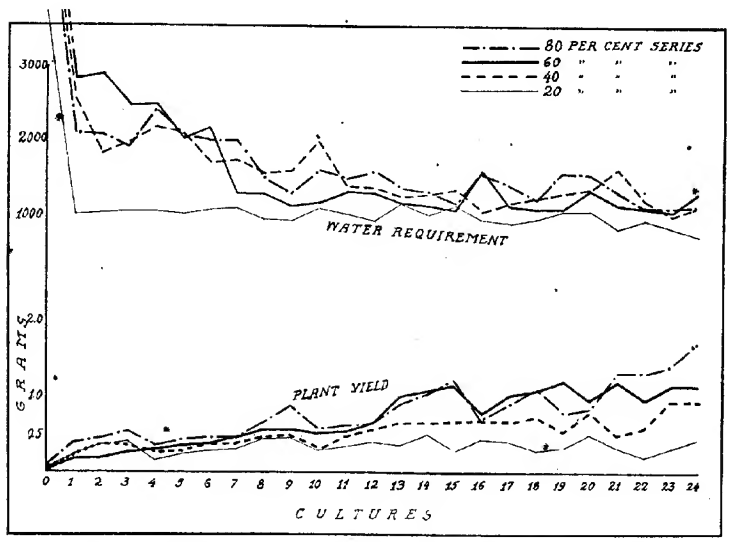

FIG. 1. THE VARIATIONS IN THE PLANT YIELD OF SEA SAND CULTURES

Kept at different moisture contents and subjected to different fertilizer treatments, and the variations in the water requirement of plants under the same conditions; the different series of cultures were kept at the indicated percentages of the water-holding capacity

yield of the 60 per cent series is well above that of the 40 per cent series. The 80 per cent series was somewhat irregular in this respect. At times only did it exceed the corresponding values for the 60 per cent series. In the last three duplicate cultures with ammonium sulfate for the nitrogenous material the difference between pots of these two series was considerable and in favor of the 80 per cent series.

Another outstanding feature of figure 1 is the relation of plant growth to the water requirement of plants. Taking it as a whole, there is a tendency for the curves of top yields to go upward, and for the curves of water requirement to go proportionally downward. There are, of course, some irregularities and excep-

tions to this general tendency, but for the average of all series this inclination can easily be traced.

The surface of the sand cultures was open and, for this reason, no account was taken of the evaporation of water. Therefore, the transpiration of plants and the evaporation of water from the surface of sand in the pots were taken together in calculating the water requirement of plants. It is fully realized that these values do not represent exactly the water requirement of plants, for which only the transpiration through the plants is usually taken. But this procedure, perhaps, more nearly approaches the field conditions, where one deals with the total water loss from the soil, as influenced by both the intake of water by the roots of the plants and the surface evaporation.

In the series with moisture equal to 20 per cent of water-holding capacity, or 4.86 per cent of water based on air-dry sand, the growth of the plants was very small, regardless of the fertilizer treatment. The plants were stunted. The variations in the dry weight of six plants were from 0.1895 gm. in culture 204 to 0.5195 gm. in culture 220. The water requirement of the tops, or the grams of water transpired and evaporated per gram of dry matter produced, varied from 823 gm. in culture 221 to 1207 gm. in culture 213. In the series whose moisture content was equivalent to 40 per cent of the water-holding capacity the variations were greater, the maximum growth being 0.9613 gm. in culture 423 and the minimum, 0.2645 gm. in pot 401. The maximum and the minimum water requirements in this series corresponded with the minimum and the maximum growth of tops, respectively. That is to say, the plants with the best growth of tops had the lowest water requirement, and vice versa. The maximum plant growth in the series with 60 per cent of the water-holding capacity was 1.2115 gm. in culture 619, and the minimum growth 0.1975 gm. in culture 602. The last culture was the one with the highest water requirement, the figure being 2901 gm., while the lowest water requirement was in culture 623 with a value of 1082 gm. In the 80 per cent series the lowest production coincided with the highest water requirement, and the highest production with the lowest water requirement. In the first case it was culture 804 with 0.3637 gm. for the dry weight of the tops and 2440 gm. for the water requirement. In the second case it was culture 824, and the figures were 1.7315 gm. and 1114 gm., respectively.

The growth of plants from cultures 1 to 12 was small in all series, and the differences between two consecutive series inconsiderable. By consulting table 2, one notices that the backward growth was merely due to the insufficient supply of nitrogen.

One can trace further the relation of lime to the total yield of plants. The culture numbers 1, 4, 7, 10, 13, 16, 19, and 22 had the smallest application of calcium carbonate. In many cases, these pots were the lowest in dry matter produced as compared with the pots with the larger application of calcium carbonate, otherwise having been treated alike.

There was some response to the larger application of monopotassium phosphate, although the degree of the response was often very low.

The plants with 0.4 gm. of ammonium sulfate per pot had a tendency to lodge. It was not entirely due to the heavy growth, because the same general phenomena were noticed in every series, though the plants in the 20 per cent series even with this application of nitrogenous fertilizer had much poorer growth than those of other series with the lesser nitrogen supply. It is supposed that this was due to the poor balance of different components in the resultant nutrient solution. As the results of subsequent experiments show (see table 10 and the accompanied discussion), this supposition was perhaps correct.

It would not be out of place at this time to discuss briefly the behavior of ammonium sulfate in comparison with nitrate of soda, or calcium nitrate, as a source of nitrogen in this experiment. In the three series out of four there was always one or more cultures that exceed both Lipman's and Shive's cultures of the same series. Only the 60 per cent series failed in this respect. Both Lipman's and Shive's cultures in this series had better yields than any pots in the same series, having ammonium sulfate for the nitrogen supply. The comparison between these nitrogenous fertilizers was not made on a strictly equal basis. But it was not intended to study the efficiency of these different nitrogenous fertilizers for plant growth. It was merely used for comparison of ammonium sulfate under different moisture conditions with the formula of known behavior in the vegctative experiments.

The untreated pots were in every case the poorest in the yield of dry matter produced in that series. After transplanting the seedlings into the pots, the plants made hardly any growth at all. It seems that the seedlings could thrive only as long as the plant-food remained in the seed. The difference in moisture content did not cause any difference in their action.

EFFECT OF MOISTURE CONTENT OF SASSAFRAS LIGHT SANDY LOAM ON THE GROWTH OF BARLEY, WITH AMMONIUM SULFATE AS THE SOURCE OF NITROGEN

Procedure

The experiment which was described in the preceding pages, was repeated is a general way with the Sassafras light sandy loam for the medium of growth The plants were grown in the same pots, 2 kgm. of soil being used. In this instance the seeds were planted directly in the pots where they were to grow. Fifteen seeds were thus planted for the purpose, and, when germinated, they were thinned to 8 seedlings per pot. During the germination the soil moisture was kept at 60 per cent of the water-holding capacity. After thinning, the water was allowed to evaporate until it reached the required per cent. From this point on it was kept constant in the way previously described for the sea sand.

The water-holding capacity of the soil was 27.7 per cent, as determined by the funnel method. Table 4 gives the per cent of moisture at which the dif-

ferent series of pots were kept and also the total amount of water maintained in the pots of each series. The hygroscopic moisture of the soil is 0.94 per cent, based on oven-dry soil.

The fertilizer treatment of the different pots was precisely the same as given in table 2 for the sea sand. Aside from the fact that some plant-food was available in the soil itself, all other relations were maintained in the same manner as in the case of the sea sand in the foregoing experiment.

The plants were harvested after 30 days of growth, beginning with the day when the moisture content of every series was brought to the required level. The results are presented in table 5 and illustrated with figure 2, following the procedure adopted for the experiment described above.

Results and discussion

Looking over table 5 and figure 2 and comparing them with table 3 and figure 1, one notices a considerable similarity between these two sets of values.

TABLE 4

Moisture content in Sassafras light sandy loam of different series

PER CENT OF WATER-HOLDING CAPACITY	PER CENT OF WATER BASED ON DRY SOIL	AMOUNT OF WATER PER POT
		cc.
20	5.54	110.8
40	11.08	221.6
60	16.62	332.4
80	22.16	443.2

Yet there are some differences which occur rather too consistently to be attributed to experimental error. The crop-yield curves of the different series run in the following order. The curve of the 20 per cent series is conspicuously low. The curves of the 40 and the 80 per cent series run fairly well together, while that of the 60 per cent series in most cases exceeds them all. In other words, the crop yield increased with the increase in the moisture content up to 60 per cent of the water-holding capacity, and then decreased with the further increase in the moisture. In this case the behavior of plants in the soil is somewhat different from that in the sea sand. It seems that the air supply is the cause of this phenomenon. In the sea sand, which naturally is more easily accessible to currents of air, the lack of aeration was not felt by the plants. In the soil, however, the aeration was somewhat slower, though the total air space remained greater than in the sand.

Another difference in the results of this experiment as compared with that of the sea sand lies in the response of plants to the application of lime. In most cases in the last three series, when nitrogen, potash and phosphorus remained the same, the smaller application of calcium carbonate gave as good a yield of barley as did the larger application. Indeed, in many cases the larger applications of calcium carbonate decreased the yield. A glance at figure 2 reveals this relation very clearly.

TABLE 5

Dry weight of barley grown in Sassafras light sandy loam of different moisture contents, total water loss per pot, and amount of water required to produce 1 gm. of dry matter under different treatments of salts

POT NO.	DRY WEIGHT OF TOPS			TRANSPIRATION AND EVAPORATION			WATER REQUIREMENT
	A	B	Average	A	B	Average	Average
Series of 20 per cent of water-holding capacity							
	gm.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*
200	0.3330	0.3430	0.3380	483	464	474	1,402
201	0.3450	0.4400	0.3925	480	529	505	1,287
202	0.5425	0.4353	0.4889	570	525	548	1,120
203	0.5635	0.3190	0.4413	539	478	509	1,100
204	0.5460	0.6525	0.5993	505	538	522	871
205	0.6500	0.5160	0.5830	529	497	513	880
206	0.7595	0.6140	0.6868	520	535	528	769
207	0.5760	0.5010	0.5385	552	487	520	966
208	0.5100	0.4645	0.4873	512	530	521	1,070
209	0.7050	0.4240	0.5645	579	529	554	982
210	0.7680	0.6720	0.7200	491	502	497	691
211	0.8980	0.6370	0.7675	544	505	525	684
212	0.5853	0.7338	0.6596	464	535	500	558
213	0.6110	0.5427	0.5769	485	522	504	873
214	0.3515	0.4837	0.4676	493	471	482	1,030
215	0.6940	0.5380	0.6160	548	531	540	877
216	0.9390	0.8560	0.8975	529	543	536	598
217	1.0580	0.9370	0.9975	548	537	543	545
218	0.8195	0.9885	0.9040	531	572	552	610
219	0.5825	0.4750	0.5288	501	461	481	909
220	0.5820	0.5795	0.5808	488	501	495	852
221	0.4030	1.1745	0.7888	464	655	560	710
222	0.9708	1.0268	0.9988	532	520	526	527
223	1.0248	0.9997	1.0123	541	524	533	527
224	1.0248	1.2330	1.1289	554	606	580	514
20S	0.8710	0.7400	0.8055	546	491	519	645
20L	0.7875	0.6802	0.7339	505	491	498	679
Series of 40 per cent of water-holding capacity							
400	0.7175	0.6440	0.6808	961	853	907	1,332
401	1.0435	1.0350	1.0395	949	989	969	933
402	1.1090	1.1860	1.1475	957	971	964	855
403	1.0235	1.1050	1.0643	990	907	949	892
404	1.1635	1.2190	1.1913	992	1,028	1,010	923
405	1.2520	1.0730	1.1625	999	927	963	829
406	1.0370	1.1080	1.0725	906	864	885	825
407	1.3050	1.3045	1.3048	983	918	951	728
408	1.2070	1.3100	1.2585	959	956	958	761
409	1.3220	1.2230	1.2725	1,003	874	939	738
410	1.3775	1.4740	1.4258	976	947	962	674

TABLE 5—*Continued*

POT NO.	DRY WEIGHT OF TOPS			TRANSPIRATION AND EVAPORATION			WATER REQUIREMENT
	A	B	Average	A	B	Average	Average
	gm.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*
411	1.1700	1.4020	1.2860	980	893	937	728
412	1.4040	1.5990	1.5015	959	981	970	646
413	1.6050	1.6760	1.6405	998	963	981	598
414	1.6515	1.3265	1.4890	1,037	894	966	649
415	1.1615	1.2405	1.2010	890	792	841	700
416	1.7795	1.7395	1.7595	1,003	1,009	1,006	571
417	1.5590	1.8030	1.6810	1,044	994	1,019	607
418	1.3280	1.8150	1.5715	905	1,037	971	618
419	1.9420	2.0805	2.0113	1,069	1,013	1,041	518
420	2.0040	2.1850	2.0945	1,084	1,072	1,078	515
421	1.8335	2.0245	1.9290	1,034	1,020	1,027	532
422	2.1600	2.2327	2.1964	1,046	1,037	1,042	474
423	2.1625	1.9178	2.0402	1,079	951	1,015	498
424	2.1550	2.0510	2.1030	1,030	1,056	1,043	496
40S	1.2800	1.6970	1.4885	968	975	972	653
40L	1.9880	1.9980	1.9930	1,060	1,084	1,072	538
Series of 60 per cent of water-holding capacity							
600	0.7920	0.6540	0.7230	1,100	998	1,049	1,445
601	1.0405	1.1150	1.0778	1,156	1,136	1,146	1,062
602	1.1965	1.1525	1.1745	1,159	1,176	1,168	997
603	1.0718	1.1540	1.1129	991	1,090	1,041	934
604	1.1110	1.2310	1.1710	1,218	1,139	1,179	1,006
605	1.0475	1.1825	1.1150	1,123	1,167	1,145	1,027
606	1.1620	1.1610	1.1610	1,097	1,065	1,081	931
607	1.3200	1.5485	1.4343	1,064	1,216	1,140	795
608	1.3635	1.3100	1.3368	1,150	1,217	1,184	886
609	1.3140	1.3335	1.3438	1,081	1,078	1,079	803
610	1.3770	1.4245	1.4008	1,113	1,088	1,101	786
611	1.3500	1.3830	1.3665	1,148	1,138	1,143	837
612	1.4540	1.2660	1.3600	1,143	1,030	1,087	799
613	1.8585	1.6940	1.7763	1,191	1,247	1,219	687
614	1.5980	1.2960	1.4470	1,142	1,082	1,112	768
615	1.5675	1.6820	1.6248	1,145	1,228	1,187	730
616	2.0680	1.8975	1.9828	1,293	1,115	1,204	607
617	1.9200	1.9050	1.9125	1,282	1,220	1,251	654
618	1.9310	1.8875	1.9093	1,275	1,179	1,227	643
619	2.2240	2.4495	2.3368	1,353	1,470	1,412	604
620	2.1815	2.5025	2.3420	1,287	1,517	1,402	599
621	2.3350	2.2715	2.3033	1,441	1,408	1,425	619
622	2.5975	2.6245	2.6110	1,433	1,557	1,495	573
623	2.4020	2.4695	2.4358	1,506	1,335	1,421	583
624	2.5230	2.4092	2.4661	1,413	1,357	1,385	562
60S	1.9780	1.8600	1.9190	1,459	1,224	1,342	699
60L	2.3820	2.7240	2.5530	1,632	1,731	1,682	658

TABLE 5—*Concluded*

POT NO.	DRY WEIGHT OF TOPS			TRANSPIRATION AND EVAPORATION			WATER REQUIREMENT
	A	B	Average	A	B	Average	Average
Series of 80 per cent of water-holding capacity							
	gm.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*
800	0.6320	0.7135	0.6728	839	907	873	1,297
801	0.9545	1.0500	1.0023	1,017	1,017	1,017	1,014
802	0.9165	1.0065	0.9615	982	991	987	1,027
803	0.9665	1.0530	1.0098	1,104	1,113	1,109	1,098
804	0.9985	0.9685	0.9835	1,031	999	1,015	1,032
805	0.9547	0.8880	0.9214	1,026	980	1,003	1,090
806	0.9210	1.0085	0.9648	1,033	1,023	1,028	1,065
807	1.1420	1.3005	1.2213	1,033	1,313	1,173	961
808	1.1250	1.3420	1.2335	1,115	1,213	1,164	944
809	1.1775	1.2470	1.2123	1,183	1,227	1,205	995
810	1.0172	1.2550	1.1361	1,164	960	1,012	979
811	1.1920	1.1695	1.1808	1,149	1,059	1,104	935
812	1.1075	1.1800	1.1438	1,108	1,101	1,105	965
813	1.7750	1.6610	1.7180	1,285	1,279	1,282	946
814	1.6450	1.7190	1.6820	1,294	1,426	1,360	809
815		1.5795	1.5795	1,162	1,344	1,344	793
816	1.5780	1.6670	1.6225	1,255	1,222	1,239	764
817	1.7100	1.5270	1.6185	1,334	1,112	1,223	756
818	1.6230	1.6830	1.6530	1,416	1,284	1,350	816
819	1.9720	2.1955	2.0838	1,450	1,587	1,519	729
820	2.1470	2.0225	2.0348	1,560	1,603	1,582	777
821	1.9310		1.9310	1,565	1,393	1,479	845
822	1.9328	1.8635	1.8982	1,398	1,311	1,355	714
823	2.0530	2.2350	2.1440	1,497	1,601	1,549	722
824	2.1655	1.9000	2.0328	1,592	1,441	1,517	746
80S	1.5695	1.4655	1.5175	1,699	1,707	1,703	1,122
80L	1.7460	1.8925	1.8193	1,523	1,707	1,615	890

All curves tend to go upward, thus showing the increasing response to the application of nitrogen. With 0.2-gm. and 0.4-gm. applications of ammonium sulfate per pot there is some difference in plant growth with the different applications of monopotassium phosphate, the greater yield being produced with the large application of this salt. This tendency is noticeable with every moisture content. No such inclination can be traced when ammonium sulfate was added only to the extent of 0.05 gm. or 0.1 gm. per pot. The limiting factor in the first 12 pots, common in all series, was nitrogen. This fact, perhaps, accounts for the lack of appreciable influence of either calcium carbonate or monopotassium phosphate.

The relation of the total yield of barley to the water requirement of plants is very apparent. With the increase in total yield there is a decrease in the amount of water required to grow a unit of dry matter. This is true in every series, but it does not hold true when one compares the cultures on the basis of

moisture content. The curve for the values of water requirement of the 20 per cent series is very erratic. The remaining three series are quite consistent in their behavior. Taking into consideration only these three series, the greatest amount of water needed to produce a unit of dry matter is noticed in the 80 per cent series; this is followed by the 60 per cent series; and the 40 per cent series is the most economical in the use of water.

In the 20 and the 40 per cent series the cultures of the minimum yield correspond with the highest water requirement of plants, and the cultures of the

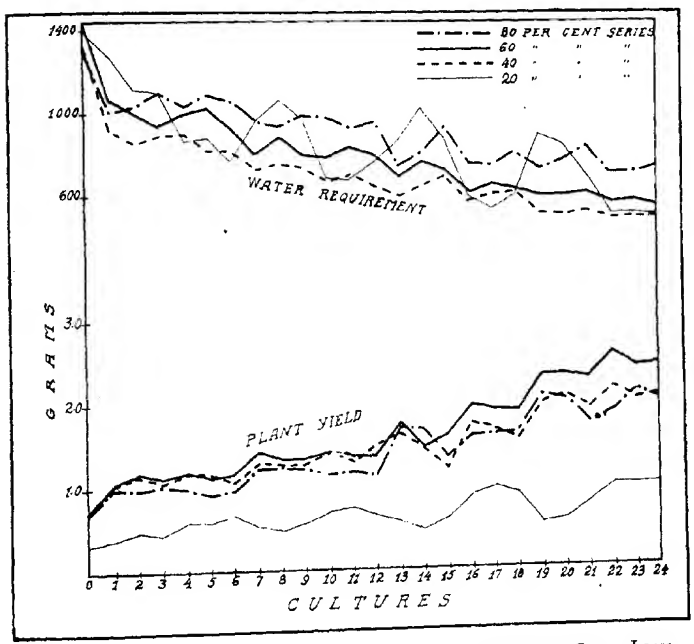

FIG. 2. THE VARIATIONS IN THE PLANT YIELD OF SASSAFRAS LIGHT SANDY LOAM

Kept at different moisture contents and subjected to different fertilizer treatments, and the variations in the water requirement of plants under the same conditions; the different series of cultures were kept at the indicated percentages of water-holding capacity

maximum yield coincide with the lowest water requirement. In the 60 per cent series the culture of the minimum yield has the highest water requirement, while the culture of the maximum yield is the second from the lowest in the water requirement. In the 80 per cent series the coincidence of lowest yield with the highest water requirement is not so exact, although quite similar.

Taking all the series together, there is a general tendency for the crop yield curves to go upward, beginning with culture 1, while for the curves of water

requirement of the plants the tendency is to go proportionally downward. This relation is quite apparent from figure 2.

The control pots, or the pots which remained untreated, had in every case the lowest yield of the series. Of the control cultures, those of the 20 per cent series had the lowest yield, while the pots of the remaining three series had practically the same yield (see no. 1 in figure 2). It seems that, beginning with the moisture content of 40 per cent of the water-holding capacity, the limiting factor was neither water nor aeration, but plant-food; for, beginning with culture 1, there was some response to the application of either one salt or another.

The standard cultures, which were represented by Lipman's (136) and Shive's (206) culture formulas, were designated by letters L and S, respectively, prefixed to the culture numbers. In the plant yields they followed the general rule for cultures with ammonium sulfate. The highest yield for both standard cultures was with the 60 per cent moisture, followed by 40 and 80 per cent in close succession, while the yield of the 20 per cent series was the lowest. In every series there were one or more cultures which exceeded in yield both the Lipman and the Shive cultures.

THE OSMOTIC CONCENTRATION OF THE SOIL SOLUTION, AND ITS EFFECT ON THE GROWTH OF BARLEY PLANTS IN SEA SAND AND SASSAFRAS LIGHT SANDY LOAM

Refering again to the figures 1 and 2, one observes that the curves for the values of the 20 per cent series stand all by themselves. This is especially true in the case of the Sassafras light sandy loam, and also in the sea sand, where nitrogen was supplied in abundance.

The question naturally arises: Why was there so great a difference in plant growth between the 20 per cent and the 40 per cent series in comparison with the differences between the 40 and 60 per cent series, and, again, between the 60 and 80 per cent series?

The answer that suggests itself is that it might be due to the increase in osmotic pressure of the solution in which the plants are grown. In preparing the cultures of the different series the same amount of salts was taken for the corresponding number of each series. The solution for the 40 per cent series, therefore, was less concentrated than that for the 20 per cent series. The solution of the 60 per cent series was less concentrated than that of the 40 per cent series; and, finally, the solution of the 80 per cent series was the most dilute of the four series. This natural progress in dilution of the nutrient solution takes place in a liquid medium, or in the homogenous system. Shall it proceed in the same way, when a solid material is introduced into the system? In other words, shall the increase in concentration in the soil solution due to the application of the nutrient solution be proportional to the decrease in the total moisture content of the soil or sand?

TABLE 6

Osmotic concentration of nutrient solutions, the sea sand treated with these solutions and the sand at the end of the vegetation experiment

	Freezing-point depression of sand treated with the nutrient solutions (before growing crop)			Osmotic concentration	Freezing-point depression of sand treated with the nutrient solutions (after growing crop)			Osmotic concentration	Freezing-point depression of the original nutrient solution			Osmotic concentration
	°C. 1	°C. 2	°C. average	Atmospheres average	°C. 1	°C. 2	°C. average	Atmospheres average	°C. 1	°C. 2	°C. average	Atmospheres average
Series of 20 per cent of water-holding capacity												
200	0.058	0.050	0.054	0.651	0.010	0.011	0.011	0.133				
201	0.340	0.342	0.341	4.100	0.093	0.078	0.086	1.037	0.165	0.169	0.167	2.014
202	0.345	0.321	0.333	4.014	0.083	0.083	0.083	1.001				
203	0.300	0.305	0.303	3.652	0.085	0.093	0.089	1.084				
207	0.363	0.320	0.342	4.122	0.111	0.101	0.106	1.278	0.185	0.191	0.188	2.267
208	0.348	0.332	0.340	4.098	0.102	0.108	0.105	1.266				
209	0.323	0.335	0.329	3.966	0.110	0.100	0.105	1.266				
213	0.508	0.503	0.506	6.097	0.158	0.152	0.155	1.869	0.223	0.223	0.223	2.688
214	0.495	0.507	0.501	6.037	0.160	0.152	0.156	1.881				
215	0.400	0.405	0.403	4.857	0.143	0.143	0.143	1.724				
219	0.510	0.520	0.515	6.205	0.201	0.197	0.199	2.400	0.282	0.284	0.283	3.401
220	0.500	0.472	0.486	5.856	0.220	0.211	0.216	2.604				
221	0.485	0.510	0.496	5.976	0.162	0.167	0.165	1.990				
Series of 40 per cent of water-holding capacity												
400	0.030	0.030	0.030	0.362	0.008	0.008	0.008	0.096				
401	0.102	0.101	0.102	1.230	0.042	0.032	0.037	0.446	0.105	0.101	0.103	1.242
402	0.104	0.102	0.103	1.242	0.030	0.028	0.029	0.350				
403	0.102	0.102	0.102	1.230	0.027	0.038	0.033	0.398				
407	0.150	0.143	0.147	1.772	0.052	0.052	0.052	0.627	0.117	0.117	0.117	1.411
408	0.128	0.148	0.138	1.664	0.029	0.030	0.030	0.362				
409	0.142	0.130	0.136	1.640	0.047	0.040	0.044	0.530				
413	0.165	0.170	0.168	2.026	0.037	0.037	0.037	0.446	0.123	0.125	0.124	1.495
414	0.165	0.158	0.162	1.954	0.057	0.055	0.056	0.675				
415	0.162	0.155	0.158	1.906	0.056	0.055	0.056	0.675				
419	0.217	0.217	0.217	2.616	0.098	0.108	0.103	1.242	0.148	0.149	0.149	1.797
420	0.222	0.197	0.209	2.520	0.075	0.075	0.075	0.904				
421	0.222	0.222	0.222	2.676	0.088	0.113	0.101	1.218				
Series of 60 per cent of water-holding capacity												
600	0.014	0.012	0.013	0.157								
601	0.077	0.075	0.076	0.916	0.054	0.055	0.055	0.663	0.050	0.050	0.050	0.603
602	0.072	0.072	0.072	0.868	0.052	0.060	0.056	0.675				
603	0.068	0.067	0.068	0.820	0.062	0.062	0.062	0.748				
607	0.080	0.081	0.080	0.965	0.036	0.034	0.035	0.422	0.065	0.065	0.065	0.784
608	0.084	0.085	0.085	1.025	0.034	0.032	0.033	0.398				
609	0.080	0.087	0.084	1.013	0.049	0.032	0.036	0.434				
613	0.129	0.124	0.126	1.519	0.070	0.067	0.069	0.832	0.080	0.080	0.080	0.955

TABLE 6—*Concluded*

	Freezing-point depression of sand treated with the nutrient solutions (before growing crop)			Osmotic concentration	Freezing-point depression of sand treated with the nutrient solutions (after growing crop)			Osmotic concentration	Freezing-point depression of the original nutrient solution			Osmotic concentration
	°C. 1	°C. 2	°C. average	Atmospheres average	°C. 1	°C. 2	°C. average	Atmosphere average	°C. 1	°C. 2	°C. average	Atmospheres average
Series of 60 per cent of water-holding capacity—Continued												
614	0.126	0.126	0.126	1.519	0.052	0.052	0.052	0.627				
615	0.126	0.122	0.124	1.495	0.062	0.061	0.062	0.748				
619	0.140	0.152	0.146	1.760	0.058	0.054	0.056	0.675	0.100	0.100	0.100	1.206
620	0.122	0.127	0.125	1.507	0.070	0.057	0.064	0.772				
621	0.129	0.127	0.128	1.434	0.057	0.057	0.057	0.687				
Series of 80 per cent of water-holding capacity												
800	0.006	0.004	0.005	0.060	0.003	0.004	0.004	0.048				
801	0.053	0.059	0.056	0.675	0.035	0.025	0.030	0.362	0.043	0.043	0.043	0.518
802	0.057	0.057	0.057	0.687	0.035	0.032	0.034	0.410				
803	0.055	0.055	0.055	0.663	0.045	0.039	0.042	0.504				
807	0.075	0.068	0.072	0.868	0.035	0.035	0.035	0.422	0.051	0.053	0.052	0.627
808	0.075	0.073	0.074	0.892	0.033	0.031	0.032	0.386				
809	0.073	0.070	0.072	0.868	0.021	0.021	0.021	0.253				
813	0.080	0.078	0.079	0.953	0.028	0.028	0.028	0.338	0.065	0.065	0.065	0.784
814	0.077	0.070	0.074	0.892	0.030	0.024	0.027	0.326				
815	0.069	0.073	0.071	0.856	0.026	0.028	0.027	0.326				
819	0.110	0.101	0.106	1.278	0.025	0.025	0.025	0.301	0.080	0.080	0.080	0.965
820	0.101	0.090	0.096	1.158	0.030	0.026	0.028	0.334				
821	0.083	0.098	0.091	1.097	0.030	0.027	0.029	0.350				

Procedure

In order to answer these questions the osmotic concentration of the nutrient solutions before and after adding them to the sand or soil was determined by means of the freezing-point method. The solutions themselves were tested in the usual way by the cryoscopic method; and those in the sand or soil by the modification of the cryoscopic method, as outlined by Bouyoucos and McCool (28). The calculations were made with the aid of the table of Harris and Gortner (86). At the end of the vegetation experiment, immediately after harvesting the plants, the soil or the sand of the pots was brought to the original moisture content. Then the contents of the pot were emptied on a clean oil-cloth, the soil was thoroughly mixed and the samples were taken to test the osmotic concentration of the material. To determine the concentration of the soil solution before growing the plants the following procedure was adopted. Twenty-five grams of the soil treated with the proper amount of calcium carbonate was taken, mixed with the required amount of the nutrient solution, and placed into the freezing test tube. On the following day

TABLE 7

Osmotic concentration of nutrient solutions, the Sassafras light sandy loam treated with these solutions, and the same loam at the end of the vegetation experiment

	FREEZING-POINT DEPRESSION OF SOIL TREATED WITH NUTRIENT SOLUTION (BEFORE GROWING CROP)			OSMOTIC CONCENTRATION	FREEZING-POINT DEPRESSION OF SOIL TREATED WITH NUTRIENT SOLUTION (AFTER GROWING CROP)			OSMOTIC CONCENTRATION	FREEZING-POINT DEPRESSION OF THE ORIGINAL NUTRIENT SOLUTION			OSMOTIC CONCENTRATION
	°C. 1	°C. 2	°C. average	Atmospheres average	°C. 1	°C. 2	°C. average	Atmospheres average	°C. 1	°C. 2	°C. average	Atmospheres average
Series of 20 per cent of water-holding capacity												
200	0.619	0.621	0.620	7.469	0.580	0.530	0.555	6.688				
201	0.653	0.658	0.656	7.902	0.615	0.620	0.618	7.445	0.155	0.155	0.155	1.869
202	0.663	0.652	0.658	7.927	0.652	0.622	0.637	7.674				
203	0.720	0.725	0.723	8.708	0.562	0.542	0.552	6.652				
207	0.703	0.728	0.716	8.624	0.587	0.585	0.586	7.060	0.166	0.164	0.165	1.990
208	0.705	0.635	0.670	8.071	0.635	0.612	0.624	7.517				
209	0.613	0.638	0.626	7.541	0.593	0.580	0.587	7.072				
213	0.677	0.708	0.693	8.348	0.590	0.630	0.610	7.349	0.189	0.187	0.188	2.267
214	0.712	0.722	0.717	8.636	0.690	0.610	0.650	7.830				
215	0.735	0.765	0.750	9.033	0.600	0.625	0.613	7.385				
219	0.764	0.799	0.782	9.418	0.675	0.668	0.672	8.095	0.248	0.250	0.249	3.002
220	0.771	0.749	0.760	9.154	0.690	0.707	0.699	8.420				
221	0.791	0.781	0.786	9.466	0.630	0.665	0.648	7.806				
Series of 40 per cent of water-holding capacity												
400	0.119	0.124	0.122	1.471	0.108	0.107	0.108	1.303				
401	0.140	0.150	0.145	1.748	0.121	0.125	0.123	1.483	0.089	0.091	0.090	1.085
402	0.160	0.175	0.168	2.022	0.124	0.126	0.125	1.507				
403	0.147	0.170	0.159	1.918	0.119		0.119	1.435				
407	0.160	0.151	0.156	1.881	0.116	0.111	0.114	1.363	0.095	0.095	0.095	1.145
408	0.160	0.151	0.156	1.881	0.106	0.116	0.111	1.339				
409	0.150	0.149	0.150	1.809	0.111	0.101	0.106	1.278				
413	0.154	0.171	0.163	1.966	0.149	0.142	0.145	1.748	0.112	0.105	0.109	1.315
414	0.188	0.151	0.170	2.050	0.139	0.128	0.134	1.506				
415	0.163	0.146	0.155	1.869	0.112	0.107	0.110	1.327				
419	0.164	0.156	0.160	1.930	0.152	0.160	0.156	1.881	0.140	0.140	0.140	1.688
420	0.164	0.169	0.166	2.002	0.153	0.145	0.149	1.797				
421	0.166	0.161	0.164	1.978	0.123	0.139	0.131	1.470				
Series of 60 per cent of water-holding capacity												
600	0.018	0.016	0.017	0.205	0.010	0.008	0.009	0.109				
601	0.042	0.041	0.042	0.506	0.018	0.020	0.019	0.229	0.061	0.063	0.062	0.748
602	0.042	0.040	0.041	0.494	0.019	0.019	0.019	0.229				
603	0.041	0.042	0.042	0.506	0.018	0.019	0.019	0.229				
607	0.045	0.045	0.045	0.542	0.028	0.032	0.030	0.362	0.070	0.067	0.069	0.832
608	0.046	0.049	0.047	0.566	0.024	0.024	0.024	0.289				
609	0.049	0.049	0.049	0.590	0.020	0.022	0.021	0.253				
613	0.045	0.044	0.045	0.542	0.032	0.022	0.027	0.325	0.072	0.071	0.072	0.868
614	0.051	0.051	0.051	0.614	0.020	0.027	0.024	0.289				

TABLE 7—*Concluded*

	Freezing-point depression of soil treated with nutrient solution (before growing crop)			Osmotic concentration	Freezing-point depression of soil treated with nutrient solution (after growing crop)			Osmotic concentration	Freezing-point depression of the original nutrient solution			Osmotic concentration
	°C. 1	°C. 2	°C. average	Atmosphere average	°C. 1	°C. 2	°C. average	Atmosphere average	°C. 1	°C. 2	°C. average	Atmospheres average
Series of 60 per cent of water-holding capacity—Continued												
615	0.045	0.049	0.047	0.566	0.027	0.027	0.027	0.325				
619	0.062	0.060	0.061	0.736	0.037	0.040	0.039	0.470	0.101	0.107	0.104	1.254
620	0.064	0.068	0.066	0.796	0.032	0.037	0.035	0.422				
621	0.068	0.073	0.071	0.856	0.029	0.036	0.033	0.398				
Series of 80 per cent of water-holding capacity												
800	0.010	0.008	0.009	0.119	0.009	0.009	0.009	0.109				
801	0.028	0.029	0.029	0.350	0.017	0.017	0.017	0.205	0.052	0.052	0.052	0.627
802	0.028	0.023	0.026	0.313	0.015	0.017	0.016	0.193				
803	0.023	0.028	0.026	0.313	0.018	0.017	0.018	0.217				
807	0.034	0.037	0.036	0.434	0.031	0.034	0.033	0.398	0.056	0.056	0.056	0.675
808	0.040	0.042	0.041	0.494	0.032	0.035	0.034	0.410				
809	0.037	0.037	0.037	0.446	0.035	0.037	0.036	0.434				
813	0.045	0.045	0.045	0.542	0.044	0.047	0.046	0.554	0.059	0.059	0.059	0.712
814	0.040	0.043	0.042	0.506								
815	0.045	0.046	0.046	0.554	0.045	0.044	0.045	0.542				
819	0.052	0.052	0.052	0.627	0.042	0.046	0.044	0.530	0.075	0.079	0.077	0.929
820	0.057	0.058	0.058	0.700	0.042	0.042	0.042	0.506				
821	0.062	0.064	0.063	0.760	0.045	0.043	0.044	0.530				

the freezing point was determined. In this test the salt combinations used were with only one amount of monopotassium phosphate, corresponding to 0.4 gm. of the salt per pot. The results are summarized in tables 6 and 7.

Results and discussion

The results of table 6 present several interesting features. Taking the values for the osmotic concentration of the sand treated with a nutrient solution and comparing them with those for the osmotic concentration of the nutrient solution itself, one notices that in most cases the concentration of the nutrient solution increases on its addition to the sea sand. The increase, however, is not the same in all series. Indeed, this value in the 20 per cent series is many times as great as in the 60 or 80 per cent series of the same number of cultures.

The differences in the total osmotic concentrations of these sand cultures with different moisture contents are, of course, even more pronounced.

The corresponding values in table 7, which represent the osmotic concentrations of the solutions in the Sassafras light sandy loam, differ somewhat

from those of the sea sand. The chief differences consist (a) in the fact that, although the osmotic concentration of the nutrient solution in the soil, after adding to it the nutrient solution, increases in the 20 and 40 per cent series, the increase is always less than the osmotic concentration of the original soil when the moisture is brought up to the required content with distilled water. In the 60 and 80 per cent series even the total osmotic concentration becomes less than that of the original nutrient solution, when this nutrient solution is introduced into the soil. (b) The total osmotic concentration of the solution in the soil of the 20 per cent series is considerably higher than in the corresponding series of the sea sand cultures. And, for this reason, the difference between the total osmotic concentration of the soil solution and the original solution is much greater than the corresponding difference in the sea sand.

TABLE 8

Comparison between the osmotic concentration of the nutrient solution and that of the soil solution after adding the nutrient solution to the soil or sand. The values are given in atmospheres

CULTURE NO.	SERIES OF 80 PER CENT		SERIES OF 60 PER CENT		SERIES OF 40 PER CENT		SERIES OF 20 PER CENT	
	Original nutrient solution	Soil solution	Original nutrient solution	Soil solution	Original nutrient solution	Soil solution	Original nutrient solution	Soil solution
	Sassafras light sandy loam							
	atm.	*atm.*	*atm.*	*atm.*	*atm.*	*atm.*	*atm.*	*atm.*
7–3	0.627	0.325	0.748	0.502	1.085	1.896	1.869	8.179
7–9	0.675	0.460	0.832	0.566	1.145	1.857	1.990	8.079
13–15	0.712	0.534	0.868	0.574	1.315	1.962	2.267	8.672
19–21	0.929	0.696	1.254	0.796	1.688	1.970	3.002	9.346
	Sea sand							
1–3	0.518	0.675	0.603	0.868	1.242	1.230	2.014	3.929
7–9	0.627	0.840	0.784	0.989	1.411	1.688	2.267	4.062
13–15	0.784	0.904	0.965	1.507	1.495	1.966	2.688	5.664
19–21	0.965	1.194	1.206	1.494	1.797	2.604	3.401	6.049

To facilitate the study of these variations table 8 is presented in which are given the average values of osmotic concentrations of three cultures that vary only in the amount of application of calcium carbonate. Table 8 enables one to compare these values in the different series. Figure 3 may facilitate the examination and interpretation of the results.

The values in table 8 and figure 3 are expressed in atmospheres of osmotic concentration. Studying this table and diagram one finds that the difference between the nutrient solution and the sand with this solution is very small in the 80 and the 60 per cent series. It increases somewhat in the 40 per cent series, and becomes greatest in the 20 per cent series. The difference is always in favor of the sand. In the soil, on the other hand, in the 80 and 60 per cent series the difference is in favor of the solution, while in the 40 and 20 per cent

series it is again in favor of the soil. The concentration is greatest in the 20 per cent series with the largest application of the ammonium sulfate. Indeed, the osmotic concentration of both the sand and the soil is consistently increased in each of the series with the increase in application of the salt ingredients.

The results discussed in the foregoing paragraphs are due, it seems, to the phenomenon of absorption. The solid particles in the soil or sand have the property of adsorbing both the water and the ions of different salts. The degree of adsorption of the different ions of the various salts fluctuates considerably with the nature of the salt and also of the soil. The water is adsorbed perhaps in direct proportion to the relative internal surface. In the

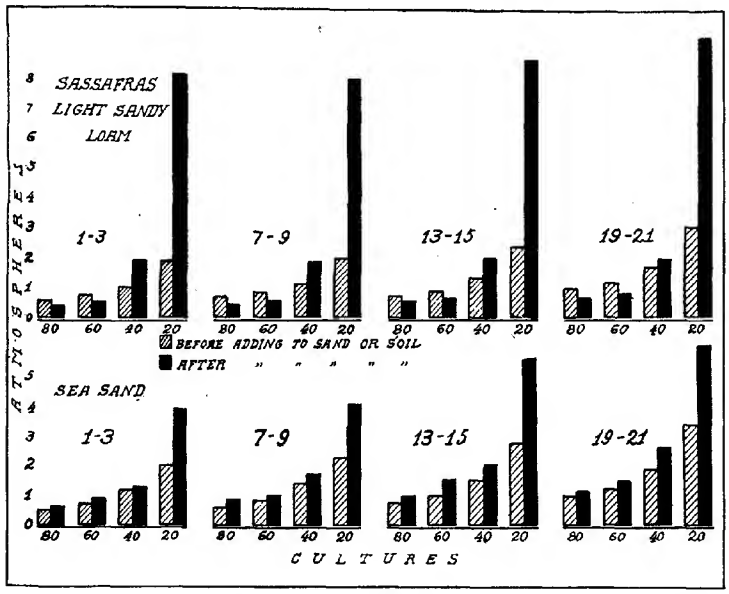

FIG. 3. COMPARISON BETWEEN THE TOTAL OSMOTIC CONCENTRATION OF THE NUTRIENT SOLUTION AND THAT OF THE SOIL OR THE SAND SOLUTION AFTER ADDING THE NUTRIENT SOLUTION TO SOIL OR SAND

The values represent the averages for 3 duplicate cultures of the same manurial treatment (with exception of calcium carbonate); 1–3, 7–9, etc. are the culture numbers; figures 80, 60, 40, and 20 represent the series of cultures kept at these percentages of the water-holding capacity

more inert solid medium, such as sand, the adsorption of salts also takes place perhaps in the proportion to the internal surface of the medium, and in proportion to the substance present in available form for reaction. In the soil, however, this relation is not so simple. There may come into play the absorptive property of the soil, which may influence the reaction very considerably. The chemical reaction, of course, is the final possibility and, undoubt-

edly, accompanies every application of the fertilizer to the soils. In the soil, besides, there may occur what is often called selective adsorption, or selective absorption, depending on the degree of reaction between the two phases. The term selective adsorption can also be applied to sand, if, on the introduction of a salt solution, one of the ions is adsorbed to a greater extent than the other, or if the amount of water adsorbed is greater than that of any of the salt ions.

Examining the values of osmotic concentration in table 8 and studying figure 3, one will notice that the phenomenon of adsorption in the sand and soil will perhaps explain the attitude of the values in different series. In the sand, on the introduction of the nutrient solution to the solid phase, some of the cations of the component salts were adsorbed to a greater extent than the anions, which resulted in the formation of acid or acids. Thus formed, the acid or acids caused an increase in the total osmotic concentration. Such a possibility was discussed in connection with some previous work (266, 267). There was, undoubtedly, some selective adsorption of water, which helped still more to increase the concentration of the resultant solution. In the 80 and 60 per cent series the influence of the adsorbed water was not sufficient to be noticeable with the method here used, because the relative amount of adsorbed water to the total water present was very small. In the 40 and 20 per cent series, however, this influence was very considerable. On reduction of the absolute volume of water, while the power of adsorption remained practically the same, the adsorbed water was, it seems, the dominant factor in modifying the osmotic concentration of the resultant solution.

The considerations presented above explain why the difference between the total osmotic concentration of the solution in the sand and the original solution is so great in every combination of the 20 per cent series.

In the soil the same agencies were at work, only differing in the degree of their action. Although the formation of an acid or acids had taken place, the total adsorption of the salts and of the acids formed in the soil was greater than the increase in concentration caused by enhanced dissociation due to acid formation. This accounts for the decrease of the concentration of the nutrient solution after adding it to the soil in the 80 and 60 per cent series. The relative adsorption of water was not great enough to modify the results considerably. On passing to the soil of the 40 per cent series, one notices that the adsorption of water begins to play a relatively important rôle. The total concentration increases rapidly. In the 20 per cent series the same phenomenon is noticed, only to a much greater degree. The salts present in the original soil, of course, add to the total osmotic concentration of the solution. The main reason for the changing osmotic concentration lies, however, in the fact that some of the water ceases to exist in the free state and the solution becomes much more concentrated. This hypothesis finds ample support in the results of recent investigations of Bouyoucos (27), who reports that some of the water, on being added to air-dry soil, becomes fixed in the latter to such an extent as to fail

to freeze when it is brought to a temperature considerably lower than the freezing point of water. The adsorption of water is greater in heavier than in lighter soils. For this reason, in the heavier types of soil one should expect a still greater osmotic concentration of the resultant solution under the same conditions, as in the 20 per cent series in the present work. In the presence of suitable substances in the soil there may take place an adsorption and also a chemical reaction that would modify the character of the resultant solution, as well as its total osmotic concentration. McCool and Wheeting (149) have found that the latter had taken place to a considerable extent, when certain single salts were introduced into the soil.

The results given in tables 6, 7, and 8 and illustrated in figure 3 throw some light on the data obtained for the growth of plants. Indeed, the values for the osmotic concentration of the series with the different moisture contents are very significant. In the 80 and 60 per cent series the absolute osmotic concentrations are quite low, and the variations are very small. These values range oinly between 0.7 and 1.5 atmospheres in the sea sand of both series. In the soil the variations were even smaller, the osmotic concentration values being here between 0.31 and 0.85 atmospheres. Within these limits the differences in plant production in cultures under different fertilizer treatments could not be attributed to the change in the osmotic, concentration of the nutrient soil solution. On the application of fertilizer in field practice the modification in osmotic concentration will not be very considerable, if the moisture content remains around 60 or 80 per cent of the saturation. If one compares, for instance, the values for the osmotic concentration of cultures 1–3, 7–9, 13–15, and 19–21 of the 80 per cent series for the Sassafras light sandy loam, one will find that on the increase in the application of ammonium sulfate from 0.05 gm. per pot to 0.4 gm. per pot the osmotic concentration increases from 0.325 atmospheres to 0.696 atmospheres, an absolute increase of only 0.371 atmospheres. Assuming a weight of 2,000,000 pounds, 0.4 gm. of ammonium sulfate corresponds to 400 pounds of the salt per acre. By doubling the amount of salt applied one would expect that the increment in the osmotic concentration of the resulting soil solution would not exceed that obtained from the first 0.4 gm. In all probability the increment would be considerably smaller, because of the increased adsorption, and also on account of the decrease in the ionic dissociation. At its maximum the osmotic concentration of a solution in the ordinary agricultural soil would not exceed perhaps 1.0–1.2 atmospheres. This osmotic concentration, it must be admitted, is not very high for the growth of plants. A higher osmotic concentration is often employed by plant physiologists (206, 226) for the best development of plants. In varying the osmotic concentration of the nutrient solution between 0.5 and 3.5 atmospheres McCall (147) finds that 2.0 atmospheres of the osmotic concentration of Shive's (206) nutrient solution, when added to the sand cultures, produces the greatest amount of dry matter of wheat. Since the results of the present work, and also of one published recently (267), show that there is, comparatively speaking, very little change taking place in the osmotic concentration of the nutrient

solution, when it is introduced into the sand at the optimum moisture content, we may ignore the difference for the present. In order to bring the osmotic concentration of our soil solution to, let us say, 2.0 atmospheres we should have to add a considerable amount of the fertilizing material. Yet, the additional amount of 0.4 gm. of ammonium sulfate per pot would bring the total amount of ammonium sulfate in the pot to 0.8 gm., which would correspond to 800 pounds of this salt per acre, an amount hardly ever used in ordinary field practice. Taking the values for the 60 per cent series the results would be somewhat higher; the difference, however, should not be very great.

From the foregoing considerations it would follow, therefore, that under field conditions, if the moisture content remains somewhere around the optimum, the ordinary application of fertilizer would not in any case modify the osmotic concentration of the soil solution to an extent sufficient markedly to affect plant growth. Increased plant production in the present experiment was evidently due to other causes. The more important among them may be (a) the increase in the total amount of plant-food, and (b) the modification in the balance of salts in the soil solution.

The osmotic concentration of the soil solution begins to play a dominant rôle when the water content becomes low. This begins to be noticeable in the 40 per cent series, in which case the osmotic concentration varies between 1.2 and 2.6 atmospheres in sea sand, and between 1.7 and 2.0 atmospheres in the Sassafras light sandy loam.

In the 20 per cent series the effects become very pronounced. The concentration in the sand varied from 3.6 to 6.2 atmospheres, and in the soil, from 7.5 to 9.4 atmospheres. At this moisture content it is a real factor, and, perhaps, is the chief reason why the yield curve of the 20 per cent series is so far below the others in the soil experiment (fig. 2). The values for the osmotic concentration of the cultures in the 40 and 20 per cent series show the importance of maintaining the proper supply of water in the soil, and seem to explain the low efficiency of some fertilizer materials in seasons of low precipitation.

The relation between the total osmotic concentration of the soil solution and plant yield can best be studied from figure 4 in which are plotted the values for the average yield of cultures 19–21, inclusive, of the Sassafras light sandy loam of the different series and also of the average osmotic concentrations for the same cultures. The treatments of cultures 19–21, as shown in table 2, differ only in the amount of calcium carbonate added. An examination of figure 4 shows in a very striking manner the dependence of plant growth on the osmotic concentration of the soil solution as brought about by the difference in the moisture content of the soil. Somewhat similar relations will be observed if parallel studies are made with the other cultures of either the Sassafras soil or the sea sand.

Refering once more to tables 6 and 7, one will notice that the osmotic concentration of the soil solution in the sand and soil was considerably reduced during the 30-day growth of barley. A greater absolute reduction in osmotic

concentration occurred in the series with the smaller moisture content. That is to say the 20 per cent series has lost most heavily, and was followed by the 40, 60, and 80 per cent series, in the order named. Comparing the same series of the two experiments, one finds that the series in the sea sand have lost more heavily, both relatively and absolutely.

EFFECT OF VARIOUS AMOUNTS OF CERTAIN NUTRIENT AND NON-NUTRIENT SALTS ON THE EFFICIENCY OF AMMONIUM SULFATE FOR THE GROWTH OF BARLEY IN SAND CULTURES

As was noticed previously in the vegetation experiments, in both the sea sand and the Sassafras light sandy loam the plants lodged in all cultures, when 0.4 gm. of ammonium sulfate was applied per pot. These cultures corresponded to culture numbers 19 to 24, inclusive. The lodging occurred in every series and, therefore, could not be attributed in every case to the heavy yield. Consulting tables 3 and 5 and figures 1 and 2, one will notice that the yield of the best pots of the 20 per cent series was not nearly as good as in the 40, 60, or 80 per cent series in cultures 13 to 18, inclusive, in which case the plants did not show any tendency to lodge. It is commonly observed in field practice that the plants usually have a tendency to lodge on land rich in available nitrogen. Although the correlation between the excessive supply of available nitrogen and the lodging of plants is very frequently noticed, yet there are times when, in the presence of an apparently excessive content of nitrogenous materials, rigid straw is produced.

Evidently, there are other factors which influence the rigidity of plant tissues. The profound influence of salt proportions on the growth of plants (226, 206, 147, 267) can not be, in seems, without some influence on the quality of their tissues. In view of this deduction it was decided to test different salts in combination with the nutrient solution, which, when present alone in the sea sand or the Sassafras light sandy loam cultures, caused the young plants to lodge.

There was another justification for performing this experiment. Although in the nutrient solution used in the present vegetative experiments there was present every element which is necessary for plant growth (according to the common conception of the subject), yet there is a possibility of increasing plant growth by some other elements or ions or salts, which may in some combinations enhance the growth of plants.

Review of literature

Common salt, sodium chloride, is usually recognized as a good amendment in soil management. Hellriegel (96) found that sodium chloride increased the yield even in the presence of an abundance of potassium. Similar results are reported by Schulze (200). Indeed, in another article the same author (199) recommends the addition of some sodium chloride with each application

of ammonium sulfate, because sodium chloride acts especially favorably with this fertilizer.

In former times calcium sulfate was often used as an amendment with some beneficial results. Yet, its value for this purpose is strongly disputed by Stebutt (213) and also by Soave (208). The latter author, however, finds that calcium sulfate produces a beneficial effect, if it is combined with soluble potash. Takeuchi (223) observed that calcium sulfate produces no beneficial effect with the acidic fertilizers and causes an increase in yield, if used with alkalinic fertilizers. He finds it to be injurious with superphosphate and ammonium sulfate, and beneficial with sodium nitrate.

Sodium silicate is found (236) to be of some benefit to crops when used in connection with green manuring.

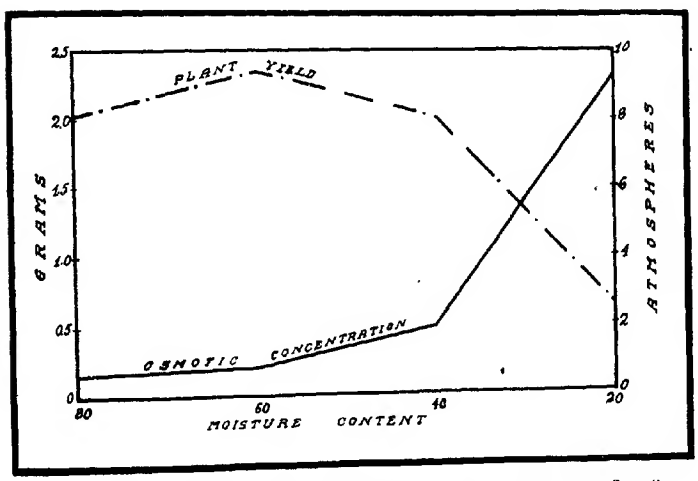

FIG. 4. THE RELATION BETWEEN THE TOTAL OSMOTIC CONCENTRATION OF THE SOIL SOLUTION AND THE PLANT YIELD IN THE SASSAFRAS LIGHT SANDY LOAM

The figures 80, 60, 40, and 20 are the percentages of the water-holding capacity of the soil

In view of the results of the investigations mentioned above and elsewhere it seemed possible that some of the substances may favorably influence the nutrient solution of the formula adopted in the present work.

Procedure

Of the two media used in previous experiments the sea sand contained the less plant-food, which was evident from the growth of barley in the untreated pots. In order better to interpret the results of the experiment, as to the influence of different salts upon the growth and development of plants, sea sand was used as the medium in this experiment. The same pots, holding 2 kgm.

of sand, were again employed. The following salts were used in addition to the formula of culture 24 (see table 2), which culture consisted of 0.4 gm. ammonium sulfate, 0.8 gm. monopotassium phosphate, 0.2 gm. magnesium sulfate, 0.05 gm. ferrous sulfate, and 2.0 gm. calcium carbonate per pot: increasing amounts of magnesium sulfate, monopotassium phosphate, and ferrous sulfate, and also different amounts of potassium chloride, sodium chloride, sodium nitrate, calcium sulfate, aluminum sulfate and sodium silicate, increasing at a uniform rate. For comparison, a culture with sodium nitrate as the source of nitrogen was employed. The ingredients used in each pot were as follows: 0.5 gm. sodium nitrate, 0.8 gm. monopotassium phosphate, 0.2 gm. magnesium sulfate, 0.05 gm. ferrous sulfate, and 2.0 gm. calcium carbonate. Table 9 gives the formula for each culture in actual application per pot, and also the calculated values for an acre 6 inches of soil (2,000,000 pounds was used for calculation).

The moisture content was kept at about 60 per cent of the water-holding capacity of the sand. The water was added daily, the pots being weighed every other day. The seeds were planted in the pots; when germinated, the seedlings were thinned to 8 for each pot. The surface of the sand was left uncovered. Thus, the evaporation of the water from the surface of the sand and the transpiration of water by the plants was taken as the total loss of water. After a 30-day growth from the time of thinning, the plants were harvested, dried and weighed. The dry weight, together with the water requirement of the plants grown in different nutrient solutions in the sand medium, are given in table 10. The same table also gives the values for the degree of stiffness of the straw. The observations to this effect were made on the day of harvesting. To represent the rigidity of the straw an arbitrary sign of small crosses was adopted. One cross (+) was taken to represent that the plants were lodged badly. Three crosses (+++) were taken to indicate that the plants stood erect, while two (++) crosses indicate medium stiffness of the straw.

In order to facilitate the examination of table 10, figure 5 is given in which the cultures with the same salts are grouped together and the results are contrasted with culture 4, or the one which received no additional salts outside of those which contained only the necessary elements. The curve for the water requirement of the plants is given in the upper portion of the diagram.

Results and Discussion

A careful examination of table 10 and figure 5 reveals the striking influence of some of the salts on the fertilizing value of the nutrient solution composed of ammonium sulfate, monopotassium sulfate, magnesium sulfate, ferrous sulfate and calcium carbonate. On addition of some of the salts to the sand cultures there was a remarkably large increase in total yield of dry matter, when this addition was made in moderate quantities. In most cases, however,

TABLE 9

Application of salts per pot containing 2 kgm. of sea sand, and the calculated values in pounds per acre

CULTURE NO.	$(NH_4)_2SO_4$		$CaCO_3$		KH_2PO_4		$MgSO_4$		$FeSO_4.7H_2O$		KCl		NaCl		$NaNO_3$		$CaSO_4$		$Al_2(SO_4)_3$		Na_2SiO_3	
	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre	Per pot	Per acre
	gm.	lbs.	gm.	lbs.	gm.	lbs.	gm.	lbs.	gm.	lbs.	gm.	lbs.	gm.	lbs.	gm.	lbs.	gm.	lbs.	gm.	lbs.	gm.	lbs.
1																						
2	0.2	200	2.0	2,000	0.4	400	0.2	200	0.05	50												
3	0.4	400	2.0	2,000	0.4	400	0.2	200	0.05	50												
4	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50												
5	0.4	400	2.0	2,000	1.2	1200	0.2	200	0.05	50												
6	0.4	400	2.0	2,000	1.6	1600	0.2	200	0.05	50												
7	0.4	400	2.0	2,000	0.8	800	0.4	400	0.05	50												
8	0.4	400	2.0	2,000	0.8	800	0.6	600	0.05	50												
9	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50	0.1	100										
10	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50	0.2	200										
11	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50	0.4	400										
12	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50			0.1	100								
13	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50			0.2	200								
14	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50			0.4	400								
15	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50					0.1	100						
16	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50					0.2	200						
17	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50					0.4	400						
18	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50							0.2	200				
19	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50							0.4	400				
20	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50							0.8	800				
21	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50									0.1	100		
22	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50									0.2	200		
23	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50									0.4	400		
24	0.4	400	2.0	2,000	0.8	800	0.2	200	0.10	100												
25	0.4	400	2.0	2,000	0.8	800	0.2	200	0.20	200												
26	0.4	400	2.0	2,000	0.8	800	0.2	200	0.40	400												
27	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50											0.1	100
28	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50											0.2	020
29	0.4	400	2.0	2,000	0.8	800	0.2	200	0.05	50											0.4	400
30	0.6	600	2.0	2,000	0.8	800	0.2	200	0.05	50												
31			2.0	2,000	0.8	800	0.2	200	0.05	50					0.5	500						

the yield reached a maximum with the quantities applied and then began to decline.

On account of differences in the influence of the different salts, it is well to consider them separately in the brief discussion of the results.

With the increase in the quantities of monopotassium phosphate (cultures 3, 4, 5 and 6) there was a slight increase in dry weight up to 0.8 gm. of the salt applied. Then some decrease resulted. With the largest application (1.6 gm.

of the salt per pot) of the phosphate the yield was even lower than with the smallest application. The results obtained with this salt could not be compared with those from other salts on an equal basis because the quantities of salts applied were different. Yet, there were evidently similar effects from monopotassium phosphate and some of the other salts.

Certain quantities of magnesium sulfate had a greater influence on the increase in plant growth than the salt discussed above.

The same thing can be said of potassium chloride, although the maximum beneficial effect in this case was passed, and was somewhere between 0.2 and 0.4 gm. of the salt per pot.

The results with sodium choride were quite in accord with those of potassium chloride, although the yield of the culture with 0.2 gm. of sodium chloride (no. 13) was the greatest in the whole series.

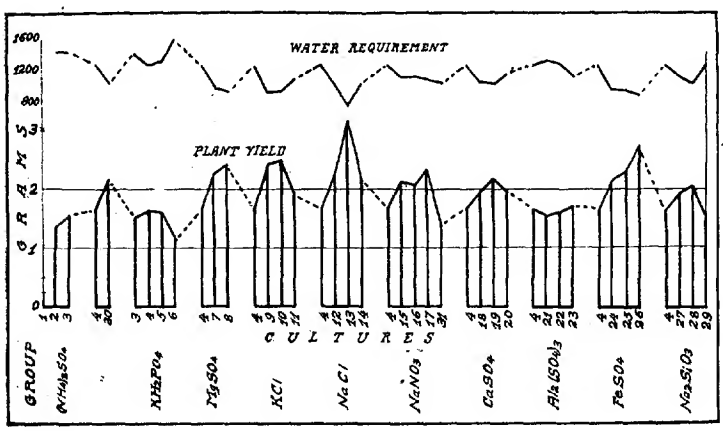

FIG. 5. THE EFFECT OF DIFFERENT AMOUNTS OF VARIOUS SALTS ON THE MANURIAL VALUE OF AMMONIUM SULFATE IN SEA SAND CULTURES, AND THE RELATION OF PLANT YIELD TO THE WATER REQUIREMENT OF PLANTS

It is intersting to note that potassium in monopotassium phosphate is present in abundance to satisfy the plant needs, yet the addition of more potassium in the form of chloride caused a considerable increase in the dry matter produced.

In the sodium chloride group there was no addition of any element that is considered essential for plant growth; nevertheless, the stimulus due to sodium chloride, in a certain combination, led to the most luxuriant growth. This could be ascribed either to the influence of sodium, of chlorine or of the combination of the two in the proper balance with other salts in the nutrient solution in the sand cultures.

The use of nitrogen in sodium nitrate, was accompanied by an increase in plant growth ($NaNO_3$) group. Either ammonium sulfate or sodium nitrate

alone gave a lower yield than the mixture of these two salts. It may be noted that 0.4 gm. of ammonium sulfate was more efficient than 0.5 gm. of sodium

TABLE 10

The dry weight of barley, the water requirement of plants, and the rigidity of the straw, as influenced by different salts in the nutrient solution in sea sand cultures; the growing period is 30 days

CULTURE NO.	YIELD OF DRY MATTER			TOTAL WATER LOSS			WATER REQUIREMENTS	RIGIDITY OF STRAW
	1	2	Average	1	2	Average	Average	Average
	gm.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	
1	0.1290	0.1190	0.1240	1,086	1,081	1,084	8,742	
2	1.3515	1.3980	1.3748	1,974	1,958	1,966	1,430	+*
3	1.6955	1.3685	1.5320	2,160	2,206	2,183	1,425	+
4	1.5985	1.7410	1.6698	2,194	2,051	2,123	1,271	+
5	1.5630	1.6605	1.6118	2,044	2,182	2,113	1,311	++
6	1.1140	1.1400	1.1270	1,836	1,804	1,820	1,615	+
7	2.6650	1.8365	2.2508	2,255	2,021	2,138	950	++
8	2.5185	2.8065	2.6625	2,158	2,253	2,206	903	+++
9	2.8600	1.9605	2.4103	2,175	2,154	2,165	899	+++
10	2.7425	2.1955	2.4690	2,380	2,147	2,264	916	+++
11	1.6155	2.1865	1.9010	2,046	2,121	2,034	1,071	++
12	2.0705	2.3755	2.2230	2,291	2,183	2,237	1,007	++
13	2.9225	3.3825	3.1525	2,332	2,288	2,310	732	+++
14	1.9885	2.2935	2.1410	2,149	2,161	2,155	1,007	++
15	2.2865	1.9615	2.1240	2,495	2,238	2,367	1,115	+
16	1.8295	2.3175	2.0735	2,347	2,489	2,418	1,166	+
17	2.2735	2.3795	2.3265	2,549	2,436	2,493	1,072	+
18	1.8525	2.0235	1.9380	2,090	1,858	1,974	1,019	++
19	2.2885	2.0595	2.1740	2,236	2,136	2,186	1,006	++
20	1.8905	1.9705	1.9305	2,214	2,254	2,234	1,159	++
21	1.6695	1.4220	1.5458	2,110	1,992	2,051	1,324	+
22	1.7985	1.4155	1.6070	2,100	2,012	2,056	1,280	+
23	1.7980	1.6385	1.7183	1,963	1,899	1,931	1,124	+
24	1.8955	2.4255	2.1655	1,957	2,086	2,022	936	++
25	2.1595	2.3910	2.2753	2,087	2,164	2,126	935	++
26	2.9815	2.5125	2.7470	2,253	2,396	2,325	846	+++
27	1.6115	2.2385	1.9250	2,119	2,221	2,170	1,127	++
28	2.2585	1.9095	2.0840	2,174	2,062	2,118	1,016	++
29	1.4665	1.5475	1.5070	1,935	1,920	1,928	1,279	+
30	1.7970	2.5740	2.1855	2,142	2,285	2,214	1,013	++
31	1.3255	1.4485	1.3870	2,147	2,352	2,250	1,622	+

* The sign of one cross (+) indicates that the plants were badly lodged; the sign of three crosses (+++) that the plants were standing erect; and the sign of two crosses (++) that the plants had straw of medium rigidity.

nitrate (see no. 4 and 31 in the sodium nitrate group, figure 5) and, therefore, nitrate of soda under these conditions was apparently benefited by the addition of ammonium sulfate more than ammonium sulfate was benefited by the addition of sodium nitrate.

The calcium sulfate group shows itself to be very similar to the potassium chloride group. Calcium in one case and potassium in another were introduced in addition to the plentiful supply of these elements in their respective cultures. The difference between these two groups consists in the fact that in the potassium chloride group there was a new ion and even element, namely, chlorine, while in the calcium sulfate group this was not the case. Both the calcium and the sulfate ions were present in some of the salts. Yet, the addition of these ions in different combination and their influence on the modification of the resultant nutrient solution was accompanied by some increase in plant growth.

An increased application of ferrous sulfate caused an increase in the production of dry matter, the largest growth having occurred with the largest application (0.4 gm).

Sodium silicate also caused an increase, when added in small amounts. There was some decrease noticed, when the application was at the rate of 0.4 gm. per pot.

Aluminum sulfate was singular in its behavior. Its effect, if there was any, was rather negative in character. There was a depressive effect with the smallest application.

Finally, with the increase in ammonium sulfate from 0.4 to 0.6 gm. per pot containing 0.8 gm. of monopotassium phosphate (pots 4 and 30) the increase in yield was greater than that from an ammonium sulfate increment ranging from 0.2 to 0.4 gm. per pot containing 0.4 gm. of monopotassium phosphate.

The nature of the results presented in table 10 and figure 5, it seems, would warrant the following general deduction. The formation of small amounts of sulfates of magnesium, calcium or iron would benefit the growth of plants like barley, if it is accompanied by the application of fertilizer represented by ammonium sulfate, superphosphate and a liberal amount of lime. The formation of these salts is possible out of the previously applied ammonium sulfate, due to the intake of the ammonium ion by plants and the leaving of some of the sulfate ion behind. The latter ion, not being able to exist as such, forms sulfuric acid, which combines with the most available bases forming sulfates. Thus, calcium, magnesium, iron and aluminum sulfates may be formed, perhaps in the order named. That is to say, if the soil contains an abundant supply of lime, some calcium sulfate will be formed, thus slightly benefiting the plant growth. In the absence of calcium carbonate some other carbonates would be used. If aluminum compounds are the most available of the bases present, the results may be entirely different. An injurious instead of a beneficial effect may be observed in that case.

On the whole, a moderate increase in the sulfate formation of certain bases in some soils, would, therefore be of considerable benefit to the crops. The increase in the growth of barley as noticed in the cultures under different salt treatments may be attributed to the modification in the balance of the component salts that go to make the soil solution. It would hardly be possible

that such an increase was caused by the increase in the osmotic concentration of the soil solution. This fact, it seems, was disclosed by the vegetation experiments described in the first part of this paper. The osmotic concentration, per se, could hardly be expected to play a very important role with these applications of the salts, unless the moisture content of the medium (sand in this case) is reduced to, or below, 40 per cent of the water-holding capacity of the medium.

The rigidity of the straw, or the ability of the plants to stand more or less erect during the first stage of growth, is another feature which was observed in connection with this experiment. An examination of the values for the stiffness of straw in table 10 shows that there is a considerable variation in the ability of plants of different cultures to stand erect. The lodging effect of the nutrient solution of culture 4 (table 10) was completely overcome by the addition of certain amounts of magnesium sulfate (no. 8), small quantities of potassium chloride (no. 9 and 10), sodium chloride (no. 13), or iron (no. 26). A considerable improvement in this direction was noticed in other cultures on the addition of small amounts of other salts.

The plants that lodged badly did not correspond with those of the heaviest yield. Indeed, the reverse tendency is noticed in the values obtained in this experiment.

Considering this feature from another point of view, the improvement of the nutrient solution of culture 4 (table 10) on the addition of some salts, such as chlorides of potassium and sodium, and sulfates of magnesium and iron, was accompanied by an increase in both plant growth and rigidity of straw. It is justifiable, therefore, to conclude that the stiffness of the straw is modified not by the presence of a definite quantity of some one element in the soil solution, but by the combination of different ingredients which constitute the soil solution. A proper balance of the salts in the nutrient solution is just as necessary for the rigidity of the straw as for the plant yield.

There is another feature which may be mentioned in connection with this experiment. A glance at the curves of figure 5 will reveal a pronounced reciprocal relation between the total yield of tops and the water requirement of plants (or the grams of water used for the growth of one gram of dry matter). In nearly every case the increase in yield was followed by a decrease in the water requirement, and vice versa. The values, on the whole, agree with those obtained in the previous experiment with sea sand kept at the same moisture content.

EFFECT OF MOISTURE CONTENT ON THE GERMINATION OF BARLEY SEEDS IN SAND AND DIFFERENT SOILS

Introduction

In the experiments on growing plants of barley in sand and soil with different moisture contents and different fertilizer treatments, the germination of seeds was allowed to take place either under very uniform conditions (sea sand ex-

periment), or under the influence of different fertilizer treatments, while the moisture content of the soil remained the same (Sassafras light sandy soil experiment). In both cases the influence of different moisture contents was eliminated as much as possible. However, since the moisture content is a very important factor in modifying the growth and development of plants, it seemed advisable to test the germination under the same conditions of moisture, under which the growth of plants had occurred.

Review of Literature

The influence of moisture content upon the germinating power of seeds has been the subject of much study. Goff (74) has observed that an excess of water, excluding the soil air, is detrimental to the germination of beet seeds. Coulter (48) made an extended study of the effect of temperature and moisture changes on the germination of seeds of different plants. As a result of his study of temperature and humidity in their relation to the germination of seeds of different grasses, Deneumostier (54) finds that different grasses require different moisture contents for optimum germination. Rye grasses, meadow fescue and tall oat grass did best when the germinating bed was kept at not less than 60 per cent of saturation, while timothy, crested dog's tail and velvet grass were injured by such moisture content. For the best germination of these the moisture content should in no case exceed 60 per cent of saturation. Livingston (139) obtained similar results. Studying *Fouquieria splendeus*, giant cactus, Mexican bean, wheat, balsam, radishes and red clover, he found that these seeds had different minimum water requirements for their germination, the variations being from 15 to 25 per cent of water of the soil studied. As a rule a somewhat better germination took place at a somewhat higher moisture content than at a minimum content.

The effect of different fertilizers and salts was also studied by a number of investigators.

De Marneffe (142) observed some differences in seed germination due to the application of fertilizers. In 1 per cent concentration, potassium sulfate and nitrate of soda were found by Hicks (99) to be detrimental to wheat, lettuce, radish and crimson clover. Phosphoric acid and lime were much less injurious in such concentrations. In an extensive study, Rusche (185) reports that when he kept his soil at 70 per cent of saturation, the application of small amounts of chlorides, nitrates, sulfates, carbonates and phosphates acted differently on the germination of seeds of the same plant. Some of the salts were injurious, others were beneficial, while the rest of them did not influence the germination to any great extent. He studied seeds of many agricultural plants and found that different crops responded differently to the fertilizer treatment.

Claudel and Crochetell (43) have found that at lower concentrations, ammonium sulfate, sodium nitrate, lime, etc. did not cause any injury in germination, but above 0.2 per cent both ammonium sulfate and sodium nitrate

resulted in injury to the seedlings. Nitrate of soda at the rate of 265 pounds per acre retarded but did not prevent germination, according to von Feilitzen (59). De Chalmont (40) also reports that a high concentration of nitrates retards the germination of seeds in water-cultures or in powdered pumice stone, while in dilute solutions they favor germination. Common salt, if present in high concentrations, is injurious to the germination of seeds, as reported by Ewart (58) and Haselhoff (92). Lime was found to increase the germinating power of a number of flower plants (31).

The reports on the effect of acids upon the germination of seeds are rather conflicting. Thus, Claudel and Crochetell (43) and Tolf (225) find that acids are injurious to germination, while Promsy (176a, 176b) reports that both organic and inorganic acids in dilute solutions favor germination.

Green manuring also may be injurious to the germination of seeds under certain conditions (62, 63, 102).

I. *Germination study in sand cultures under different fertilizer treatments*

Procedure

The first part of the germination tests of the present work was carried out in sea sand. For this purpose 300 gm. of sea sand was weighed out and placed in tumblers. The sand was supplied with exactly the same proportions of fertilizing materials and moisture as in the corresponding vegetation experiments with the same material. Ten selected barley seeds were placed about half an inch deep in the moist and compacted sand of each tumbler, the surface of the sand was again slightly compacted, and the tumblers were covered with squares of glass. This was done to prevent evaporation of water from the surface. On the seventh day from seeding the germinated seedlings were counted in each tumbler. The results are given in table 11, which show the number of seeds germinated in the cultures of each of the four series. The 20, 40, 60 and 80 per cent series contained 4.86, 9.72, 14.58, and 19.44 per cent of water, respectively, as based on the air-dry sand, whose water-holding capacity was equal to 24.3 per cent. Each series consisted of 54 tumblers.

Results and discussion

Table 11 reveals a remarkable fact. It not merely shows that the moisture content equivalent to 40 per cent of the water-holding capacity did not retard the germination, but that the moisture content equivalent to 20 per cent of the saturation was sufficient for nearly a perfect stand. The germination was strikingly uniform in all the series, the seeds of the 20 per cent series came through about 24 hours later than those in the remaining series. Only in a few tumblers, namely, in numbers 19 to 23, inclusive, of the 20 per cent series, were the seedlings more or less retarded. There were also two sets in the 40 per cent series that were somewhat behind in their coming through the sand.

Shive's (206) nutrient solution in both the 20 and 40 per cent series showed some retardation, this being more pronounced in the 20 than in the 40 per cent series. With these few exceptions there was apparently no injurious effect of the fertilizing salts applied in any combination.

TABLE 11

Number of barley seeds germinated in sea sand cultures under different fertilizer treatments in four different moisture contents, 10 seeds being planted

SERIES...	20		40		60		80	
Per cent of H_2O in sea sand	4.86		9.72		14.58		19.44	
Trial.....	1	2	1	2	1	2	1	2
No.								
0	9	10	10	9	10	9	9*	10*
1	10	10	10	10	10	10	10	10
2	10	10	10	10	10	10	10	10
3	10	10	10	10	10	10	10	10
4	8	10	10	10	10	10	10	10
5	10	10	10	10	10	10	10	10
6	10	10	10	10	10	9	10	10
7	10	10	10	10	10	10	10	10
8	10	10	10	10*	10	10	10	10
9	9	10	10	10	10	10	10	10
10	10	10	10	9	10	10	10	10
11	10	10	10	10	10	9	10	9
12	10	10	10	10	10	10	10	9
13	10	10	10	10	10	10	10	10
14	10	10	10	10	10	10	10	10
15	10	10	10	10	10	9	10	10
16	10	10	10	10	10	10	10	10
17	9	10	9	10	9	10	10	10
18	10	10	10	10	9	10	10	10
19	10*	10*	10	10	9	10	10	10
20	9*	10*	10	10	10	10	10	10
21	9*	10	10	10	10	10	10	10
22	10*	9*	10*	10*	10	10	10	10
23	10*	10*	10*	10*	10	10	10	10
24	10	10	10	10	10	10	10	10
L‡	10	10	10	9	9	10	10	10
S‡	8†	9†	10*	10*	10	10	9	10

* Germination slightly retarded.

† Germination considerably retarded.

‡ L—Lipman's; S—Shive's.

In view of the osmotic concentration values presented in table 6 and also in view of the growth of plants in sand with different moisture contents, the results of the germination test are rather interesting. The germination of seeds is evidently not affected by differences in the moisture content of the

sand to the same extent, as is the growth of plants. It seems that for the initial development of the embryo very little available water is required, and that this water can be obtained from nutrient solutions of very high osmotic concentration. By consulting table 6 one will observe that the osmotic concentration of the nutrient solutions in the sea sand of the 20 per cent series varies from 3.6 to 6.2 atmospheres. This, of course, is considerable, but, it seems, that such osmotic concentration does not prevent the seeds from obtaining the water for their hydration from the thin films surrounding the sand particles.

II. *Germination study in different types of agricultural soils*

Procedure

The results obtained with the sea sand naturally raise the question, whether different types of agricultural soils would permit the seeds to germinate under identical moisture conditions.

In order to throw some light on this question, other germination tests were made. For this purpose, the following soils, together with sea sand, were used: Sassafras light sandy loam, Sassafras medium silt loam, Elkton clay loam, and muck. Muck was procured from the fields of the Alphano Humus Company,[2] while the remaining soils were obtained from the fields of the New Jersey State Agricultural College and Experiment Stations at New Brunswick. The hygroscopic water of the air-dry soil and sand, as well as their water-holding capacity, are given in table 12. In this experiment 250 gm. of soil and 150

TABLE 12

Water-holding capacity of different soils, as based on air-dry soil, and the hygroscopic moisture of these soils

SOIL	HYGROSCOPIC MOISTURE OF THE AIR-DRY SOILS			WATER-HOLDING CAPACITY (FUNNEL METHOD)		
	1	2	Average	1	2	Average
	per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
Sea sand	0.054	0.054	0.054	24.2	24.5	24.35
Sassafras light sandy loam	0.94	0.94	0.94	27.7	27.8	27.75
Sassafras medium silty loam	1.72	1.71	1.72	51.9	53.0	52.45
Elkton clay loam	1.82	1.80	1.81	40.8	41.3	41.1
Muck	26.19	25.79	25.99	143.6	144.0	143.8*

* By Hilgard method average of two determination = 143.3 per cent.

gm. of muck were used in the tumblers. One half of the tumblers remained untreated, while the other half received a nutrient solution corresponding to the formula of no. 15 in the vegetation experiments (table 2). The moisture content of each soil varied from 10 to 80 per cent of its water-holding capacity, as based on the air-dry soil. The actual moisture content of each series, that is, the hygroscopic water plus the water added to the soil, is given in table 13.

[2] The author is indebted to Dr. H. Clay Lint for his kindness in supplying the muck soil.

Figure 6 shows the number of seeds germinated in 10 days after the planting. To complete the curve for the results with the sea sand the figures for the 40, 60 and 80 per cent series were taken from the first experiment on germination (table 11). The curves represent the values for the untreated sand or soils.

TABLE 13

Actual moisture contents of different soils of the various series in the germination experiment (Water added plus the hygroscopic moisture)

SERIES	SEA SAND	SASSAFRAS LIGHT SANDY LOAM	SASSAFRAS FINE SILTY LOAM	ELKTON CLAY LOAM	MUCK
	per cent	*per cent*	*per cent*	*per cent*	*per cent*
10	2.48	3.71	7.47	5.92	40.37
15	3.70	5.09	10.35	7.98	47.56
20	4.91	6.48	13.22	10.03	54.75
40	9.77	12.02	24.72	18.25	83.51
60	14.63	17.56	36.22	26.47	112.27
80	19.49	23.10	47.72	34.69	141.03

TABLE 14

Number of barley seeds germinated in 5 and 10 days after planting in different soils kept at different moisture contents

SERIES	10		15		20		40		60		80	
Number of days after planting	5	10	5	10	5	10	5	10	5	10	5	10
Sea sand												
Untreated	3	9	10	10	10	10						
Untreated	4	10	10	10	10	10						
Treated	3	9	9	10	10	10						
Treated	3	9	8	10	10	10						
Sassafras light sandy loam												
Untreated	0	0	0	0	0	5	10	10	5	5	5	5
Untreated	0	0	0	0	0	6	8	10	7	7	2	2
Treated	0	0	0	0	0	2	10	10	9	9	3	3
Treated	0	0	0	0	0	3	10	10	3	3	2	2
Sassafras medium silt loam												
Untreated	0	0	0	0	3	9	10	10	3	3	1	1
Untreated	0	0	0	1	2	9	10	10	4	4	2	2
Treated	0	0	0	0	2	10	10	10	1	1	0	0
Treated	0	0	0	0	3	9	10	10	2	2	0	0
Elkton clay loam												
Untreated	0	0	0	0	0	0	9	9	9	9	1	1
Untreated	0	0	0	0	0	0	7	8	6	6	0	0
Treated	0	0	0	0	0	0	9	9	7	7	4	4
Treated	0	0	0	0	0	0	8	10	7	7	1	1
Muck												
Untreated	0	0	0	0	0	0	9	9	10	10	6	8
Untreated	0	0	0	0	0	0	9	10	10	10	6	7
Treated	0	0	0	0	0	0	10	10	10	10	9	9
Treated	0	0	0	0	0	0	10	10	10	10	7	8

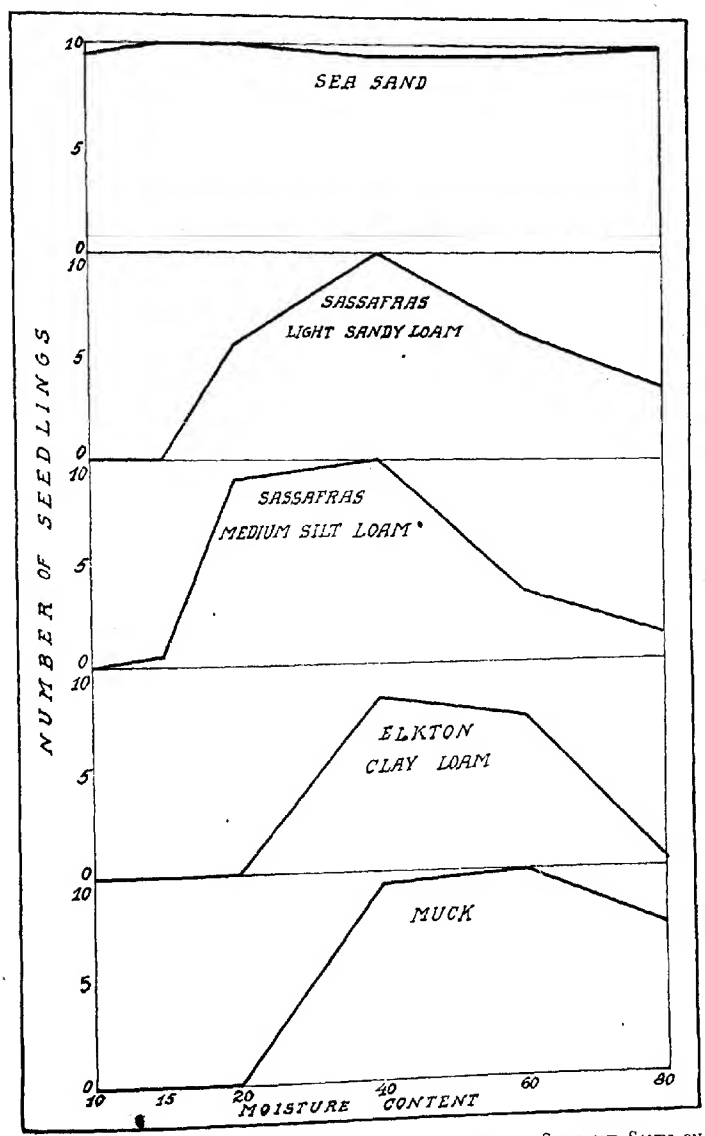

FIG. 6. THE EFFECT OF THE MOISTURE CONTENTS OF DIFFERENT SOILS AND SANDS ON THE GERMINATION OF BARLEY SEEDS

Results and discussion

The results given in table 14 and illustrated in figure 6 show very clearly that the soils differ considerably from the sand in their behavior toward moisture.

In the sea sand a nearly perfect germination was obtained with any moisture content studied, which varied from 2.48 to 19.49 per cent as based on the dry sand. In the soils, however, the curves for germination are entirely different. There was no germination in any of the soils at 10 or even 15 per cent of the water-holding capacity. In the clay and muck soils there was no germination even at 20 per cent of saturation. In both Sassafras soils a considerable germination took place at 20 per cent of the saturation, and the maximum germination was at 40 per cent. Indeed, in every soil the moisture equivalent to 40 per cent of the water-holding capacity was either the best or nearly the best for the optimum germination of barley seeds. Sixty per cent of the saturation for most of the soils was not so good as 40 per cent. In all soils without exception the moisture equivalent to 80 per cent of the saturation was very far from the optimum for the germination of the seeds.

On the whole, the sand has a much greater range in moisture for good germination of barley seeds, while in the different soils studied the range is considerably narrower. In soils with the low moisture content the failure of seeds to germinate was due, partially perhaps, to the high osmotic concentration. But the dominant factor, it seems, was the lack of a sufficient amount of free water around the seeds. That water had been taken in by the seeds was evident from the fact that, after 10 days from planting the barley, in 20 and even 15 per cent of the water-holding capacity (in several cases, in 10 per cent, as well) the seeds examined were swollen considerably. But there was not enough of the free water to start the embryo cells to multiply. On account of the internal friction the movement of water in soils is slower than in sand. In different soils the rate of the movement of water decreases with the increase in the per cent of the fine material, and the movement of the moisture in the same soil decreases with the decrease beyond certain limits in the moisture content. Besides, at the same degree of saturation the per cent of free water is undoubtedly different in different types of soil. These facts account for the variations in the behavior of the sand and soils in the present experiment.

The failure of the seeds to germinate normally in the soils having a moisture content 80 per cent of the saturation is possibly due to the poor aeration and the lack of oxygen for the metabolism of the seed tissue.

The treatment of the soils or the sand with the nutrient solution had a very limited effect, if any, on the ability of seeds to germinate. The salts used would correspond to but a normal application in farm practice. Based on pounds per acre of 2,000,000 pounds in soils and 1,000,000 pounds in muck, the application of salts would equal to 2,000 pounds of calcium carbonate, 200 pounds of ammonium sulfate, 400 pounds of monopotassium phosphate, 200 pounds of magnesium sulfate and 50 pounds of ferrous sulfate. It seems, therefore, that

a normal application of fertilizers, if these fertilizers are well incorporated in the soil, does not have any injurious effect on the germination of the seeds in question. If the fertilizer is applied in drills at the time of seeding, the soil solution immediately surrounding the seeds may become very concentrated, and, consequently, an injurious effect on seed germination may be expected. Any drought prevailing during the germination period, may still further aggravate the situation brought about by the fertilizer treatment.

SUMMARY

The experimental work described above deals with the effect of ammonium sulfate, as used in different combinations with other salts, on the germination and the growth of barley in sand and soil cultures with different moisture contents. The fertilizer treatment consisted of ammonium sulfate, monopotassium phosphate, calcium carbonate, magnesium sulfate and ferrous sulfate in different proportions. The effect of different amounts of potassium chloride, sodium chloride, sodium nitrate, calcium sulfate, aluminum sulfate, and sodium silicate on the main formula of the fertilizing treatment also was studied. The osmotic concentration of the nutrient solutions in many cases was determined both before and after the application of these solutions to the sea sand or the Sassafras light sandy loam, in which the plants were grown. The moisture content was kept more or less constant by adding water daily and weighing the pots every other day, each time bringing the moisture content to the original. The moisture of the pot cultures was kept at four different percentages: 20, 40, 60 and 80 per cent of the water-holding capacity of the sand or the soil studied. In the germination experiment, besides the four variations mentioned above, moisture contents of 10 and 15 per cent of the water-holding capacity also were included.

The results obtained in these studies may be summarized as follows:

1. The moisture content of the soil has a very marked influence on the growth and the development of plants. In the sand cultures the plant yield increases on the increase in moisture content from 20 to 80 per cent of the water-holding capacity of the sand. In the Sassafras light sandy loam the plant yield increases with the increase in moisture content up to 60 per cent of the water-holding capacity of the soil, while the further increase in moisture brought a considerable decrease in the yield of dry matter of barley. The plant growth in the soil or sand kept at a moisture content equivalent to 20 per cent of saturation was very small, and without any direct relation to the water present, as compared with the series of the higher moisture content.

2. With a constant moisture content in the sand, the plant yields increased with the increase in the application of ammonium sulfate, calcuim carbonate, or monopotassium phosphate. The response in the plant growth to the applications of these salts in the amounts used was in the order named. In the Sassafras light sandy loam a similar response to applications of nitrogen was noticed, but not to those of lime, and very little to those of phosphorus.

3. The difference in plant growth in the various moisture contents was attributed to the two factors, the total plant-food remaining the same: (a) the difference in concentration of the soil solution, and (b) the aeration of the soil.

4. The osmotic concentration of the soil solution increases with the decrease in the moisture content of sand or soil, but the changes are not proportional to one another.

5. The change in the osmotic concentration of the soil solution with the change in water content from one series to another was greater than the change in the osmotic concentration of the soil solution due to the different fertilizer treatments, if the water content remained the same.

6. In cultures with the moisture content corresponding to 80 and 60 per cent of the water-holding capacity, the osmotic concentration of the soil solution varied between 0.7 and 1.5 atmospheres in sea sand, and between 0.31 and 0.85 atmosphere in Sassafras light sandy loam. With a moisture content corresponding to 40 per cent of the water-holding capacity the corresponding values for sand were 1.2 and 2.6 atmospheres, and for soil, 1.7 and 2.0 atmospheres. With 20 per cent of the water-holding capacity the maximum and the minimum values in sand were 3.6 and 6.2 atmospheres, respectively, and in soil, 7.5 and 9.4 atmospheres.

7. The osmotic concentration of the soil solution following the normal application of a fertilizer is not great enough to influence plant growth, if the moisture content of the soil is at its optimum (about 60 per cent of saturation). It becomes an important factor only when the moisture content of the soil is considerably reduced (to 40 per cent of the saturation, or lower).

8. On adding the nutrient solution to the soil its osmotic concentration decreases, as measured by the cryoscopic method, if the moisture content of the soil is maintained at 60 or 80 per cent of its water-holding capacity. In the 40 and 20 per cent series of the water-holding capacity the osmotic concentration of the soil solution after the addition of the nutrient solution was greater than that of the nutrient solution itself. By adding the nutrient solution to the sea sand at any of these four moisture contents its osmotic concentration increased. The latter phenomenon was attributed to the formation of acid or acids with the high moisture content, and to the formation of acid or acids and the adsorption of water with the low moisture content. The adsorptive and the absorptive capacity of the soil for salts prevented the effect of these two agencies from becoming noticeable in the change of the osmotic concentration of the soil solution in the Sassafras light sandy loam with 60 and 80 per cent of the water-holding capacity.

9. The osmotic concentration of the soil solution at the end of the growing period (30 days) was smaller than at the beginning of the experiment. The decrease was greater in the sand than in the soil, and also in the lower moisture content than in the higher moisture content of either the sand or the soil.

10. The nutrient solution consisting of 0.4 gm. ammonium sulfate, 0.8 gm. monopotassium phosphate, 2.0 gm. calcium carbonate, 0.2 gm. magnesium

sulfate, and 0.05 gm. ferrous sulfate per 2 kgm. of sea sand cultures with a moisture content equivalent to 60 per cent of the water-holding capacity was benefited by the additional application of magnesium sulfate and ferrous sulfate, and also by small applications of potassium chloride, sodium chloride, sodium nitrate, calcium sulfate, and sodium silicate. The beneficial effect of these salts on plant growth was attributed to the improvement in the balance of the ions of the component salts in the resultant soil solution. Aluminum sulfate under similar conditions had caused some injury to plants.

11. The rigidity of the straw of plants was modified by different salts added to the nutrient solution. A proper balance in the nutrient solution is essential for the rigidity of the straw. The lodging effect of the large amount of nitrogenous material may be entirely subdued by modifying the proportions between the component salts in the nutritive solution in sand cultures.

12. When the evaporation of water from the surface of the sand or of the soil was taken together with the transpiration of water by plants, it was found that the water requirement of plants diminishes with the increase in plant yield, and vice versa.

13. The use of water by plants in Sassafras light sandy loam is most economical with a moisture content equivalent to 40 per cent of the water-holding capacity, and is followed by that of 60 and 80 per cent in the order named.

14. If plant-food is the limiting factor in either soil or sand, the variations in the moisture content do not affect the plant growth.

15. The germination of seeds of barley is influenced by the same general agencies that affect the growth of plants, although not to the same extent. The germination in the sea sand takes place even when the moisture content is reduced to 10 per cent of the water-holding capacity (2.48 per cent based on the dry sand), or when it is raised to 80 per cent of saturation (19.49 per cent water). In the soils (four different soils having been studied), the limits in moisture content between which the germination of seeds can normally take place is much narrower. In two soils the germination could not take place at 20 per cent of the water-holding capacity, while in no case with the soils studied did it take place below this per cent. In all of the soils studied germination was retarded at 80 per cent of the saturation. In three soils out of four the best germination was observed at 40 per cent, followed by 60 per cent of the water-holding capacity.

16. The treatment of the sand or soil with a nutrient solution at the high moisture contents had very little, if any, effect on the germination of barley seeds. With the decrease in moisture content there was noticed some retardation in seed germination, when it was accompanied by the application of nutritive salts.

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REFERENCES

(1) ANONYMOUS 1894 Field experiments with fertilizers in Great Britain. *In* Bd. Agr. and Fisheries [London], Ann. Rpt., Agr. Ed. and Research, p. 41–66.

(2) ANONYMOUS 1895 Manurial experiments in various parts of Scotland. *In* Trans. Highland and Agr. Soc. Scot., ser. 5, v. 7, p. 436–459.

(3) ANONYMOUS 1908 Seeding canes and manurial experiments at Barbados, 1906–1908. *In* Imp. Dept. Agr. West Indies Pamphlet 59, p. 101.

(4) ANONYMOUS 1909 The results of the flax experiment farms of Selesia in 1908. *In* Ztschr. Landw. Kammer Schlesien, Bd. 13, No. 40, p. 1197–1203.

(5) ANONYMOUS 1910 Tests with four nitrogenous manures. *In* Dept. Agr. and Tech. Instr. Ireland Jour., v. 10, no. 2, p. 352–354.

(6) ANONYMOUS 1911 Field experiments, 1910. *In* Dept. Agr. and Tech. Instr. Ireland Jour., v. 11, no. 2, p. 238–302.

(7) ANONYMOUS 1913. Tests with four nitrogenous manures. *In* Dept. Agr. and Tech Instr. Ireland Jour., v. 13, no. 2, p. 294–299.

(8) ANONYMOUS 1913 Effect of certain artificial manures on the bushel weight of oats. *In* Dept. Agr. and Tech. Instr. Ireland Jour., v. 13, no. 4, p. 705–707.

(9) ANONYMOUS 1915 Experiments on permanent grass land, 1915. *In* Agr. Students' Gaz., n. s., v. 17, no. 4, p. 162–165.

(10) ANSTEAD, R. D. 1911 Manurial experiments with Herea rubber. *In* Planters' Chron., v. 6, no. 45, p. 690–692.

(11) BACHMANN, H. 1901 Nitrate of soda and sulfate of ammonia. *In* Fuhling's Landw. Ztg., Bd. 50, No. 11, p. 386–387.

(12) BACHMANN, H. 1904 Further experiments with ammonium sulfate and sodium nitrate. *In* Fuhling's Landw. Ztg., Bd. 53, No. 13, p. 502–506; No. 14, p. 539–543.

(13) BACHMANN, H. 1907 Experiments with ammonium sulfate. *In* Fuhling's Landw. Ztg., Bd. 56, No. 15, p. 530–535.

(14) BACHMANN, H. 1907 On the action and residual effects of Thomas ammonium phosphate. *In* Fuhling's Landw. Ztg., Bd. 56, No. 24, p. 875–876.

(15) BACHMANN, H. 1908 Comparative tests of sulfate of ammonia and nitrate of soda. *In* Landw. Wchnbl. Schles.-Holst., Bd. 58, No. 42, p. 708–712.

(16) BACHMANN, H. 1909 Nitrogen fertilizer experiments on meadows. *In* Landw. Wchnbl. Schles.-Holst., Bd. 59, No. 2, p. 22–24.

(17) BALLARD, W. C., and VOLCK, W. H. 1914 Winter spraying with solutions of nitrate of soda. *In* Jour. Agr. Res., v. 1, no. 5, p. 437–444.

(18) BAUMANN, A., and PAUL, H. 1915 Report on field fertilizer experiments at the Bernau moor cultivation Station. *In* Mitt. Deut. Landw. Gesell., Bd. 30, No. 42, p. 627–630; No. 43, p. 651–653; No. 45, p. 686–688.

(19) BAUWENS, L. 1908 Tests of the new nitrogenous fertilizers on potatoes and turnips in 1907. *In* Jour. Soc. Cent. Agr. Belg., v. 55, no. 5, p. 169–172.

(20) BERRY, R. A. 1909 Reports on experiments with some new nitrogenous manures on oats, hay, potatoes, mangels and sugar beets. *In* West of Scot. Agr. Col. Ann. Rpt., v. 9, p. 15–31.

(21) BLAIR, A. W., AND MCLEAN, H. C. 1916 The influence of lime on the yield and nitrogen content of corn. *In* Soil Sci., v. 1, no. 5, p. 489–504.

(22) BLOBEL, E. 1908 Investigations on the action of sulfate of ammonium and nitrate of soda as fertilizers. (Untersuchungen über den Wirkungswert von schwefelsauren Ammoniak und Chilisalpeter als Dungemittel.) Inaug. Diss. Leipsig, 1908, 100 p.

(23) BOLIN, P. 1906 Cooperative fertilizer trials in Sweden, 1905. *In* K. Landtbr. Akad. Handl. och Tidskr., v. 45, Bihang, p. 121.

(24) BOLIN, P. 1907 Report of coöperative fertilizer trials, 1906. *In* K. Landtbr. Akad. Handl. och Tidskr., v. 46, Bihang, p. 103.

(25) BONOMI, Z., AND MORGRETH, G. 1914 Comparison of four nitrogenous materials as sources of nitrogen. *In* R. Lab. Chim. Agr. Udine, Ric., Sper. e. Attiv. Spiegata, v. 4, p. 29–34.

(26) BOUSSINGAULT, M. 1854 Recherches sur la végétation entreprises dans le but d'examiner si le plantes fixent dans leur organisme l'azote qui est à l'etat gaseux dans l'atmosphere. *In* Ann. Chim. et Phys., ser., 3, t. 41; p. 5–60.

(27) BOUYOUCOS, G. J. 1917 Classification and measurement of the different forms of water in the soil by means of the dilatometer method. Mich. Agr. Exp. Sta. Tech. Bul. 36.

(28) BOUYOUCOS G. J., AND MCCOOL, M. M. 1915 The freezing-point method as a new means of measuring the concentration of the soil solution directly in the soil. Mich Agr. Exp. Sta. Tech. Bul. 24.

(29) BOVELL, J. R., AND D'ALBUQUERQUE, J. P. 1897 Report on the results obtained with sugar cane on the experimental fields at Dodds Reformatory. *In* Rpt. Exp. Fields Dodds Reform. Barbados, p. 80.

(30) BOVELL, J. R., et al. 1912 Experiments with sugar cane, cotton and legumes. *In* Rpt. Local Dept. Agr. Barbados, p. 345.

(31) VON BREHMER 1907 The importance of lime as a plant food for seedlings. *In* Gartenwelt, Bd. 11, No. 14, p. 163–164.

(32) VON BREHMER 1910 Sodium nitrate as compared with ammonium sulfate. *In* Gartenwelt, Bd. 14, No. 28, p. 342–344.

(33) BROOKS, W. P. AND THOMSON, H. M. 1900 Report of the agriculturist. *In* Mass. Agr. Exp. Sta. Ann. Rpt. 1899, p. 32–35.

(34) BROOKS, W. P., et al. 1906 Report of the agriculturist. *In* Mass. Agr. Exp. Sta. Ann. Rpt. 1906, p. 23–60.

(35) BROWN, B. E. 1909 Concerning some effects of long-continued use of sodium nitrate and ammonium sulfate on the soil. *In* Penn. Agr. Exp. Sta. Ann. Rpt. 1909, p. 84–92.

(36) BROWN, B. E., et al. 1910 Soil studies in 1909. *In* Penn. Agr. Exp. Sta. Ann. Rpt., p. 47–129.

(37) BUCHNER 1910 Comparative fertilizer experiments with lime nitrogen. *In* Sachs. Landw. Ztschr., Bd. 58, No. 12, p. 154–155.

(38) CALVIN, M. V. 1911 Corn production. Ga. Agr. Exp. Sta. Bul. 93, p. 147–154.

(39) CARUSO, G. 1906 Fertilizer experiments in 1905 with calcium cyanamid in comparison with other nitrogenous fertilizers and liquid manure. *In* Atti R. Accad. Econ. Agr. Georg. Firenze, ser. 5, v. 3, no. 2, p. 228–238.

(40) DE CHALMONT, G. 1894 The influence of nitrates on germinating seeds. *In* Agr. Sci., v. 12, no. 10, 11, p. 463–464.

(41) CHODAT, R., et al. 1907 The growth of plants and the absorption of mineral substances. *In* Bul. Herb. Boissier, ser. 2, t. 7, no. 4, p. 350–352.

(42) DE CILLIS, E. 1908 Various experiments with calcium cyanamid. *In* Ist. Agr. Siciliano "Valdisavoja" Relaz., 1906–1908, p. 117–130.

(43) CLAUDEL AND CROCHETELL 1896 Influence of certain substances used as fertilizers on germination. *In* Ann. Agron., t. 22, no. 3, p. 131–142.

(44) CLAUSEN, H. 1900 Nitrate of soda and sulfate of ammonia on marsh soils. *In* Landw. Wchnbl. Schles.-Holst., Bd. 50, No. 33, p. 562–564.

(45) CLAUSEN, H. 1905 Ammonium salts or nitrate of soda. *In* Deut. Landw. Presse, Bd. 33, No. 72, p. 611, 612.

(46) CLAUSEN, H. 1908 Das Ergebnis eines umfangreichen Dungungs-Versuches auf Sandboden in schechter Kulture. *In* Deut. Landw. Presse, Bd. 35, No. 11, p 105–106; No. 12, p. 115–116.

(47) COLEBATCH, W. J. 1914 Soil water in relation to plant growth. *In* Jour. Dept. Agr. So. Aust., v. 18, no. 4, p. 363–375.

(48) COULTER, S. 1898 Notes on the germination and seedlings of certain native plants. *In* Proc. Indiana Acad. Sci., p. 215–222.

(49) CROWTHER, C. 1909 The influence of artificial manures on the yield and malting quality of barley grain. Univ. Leeds and Yorkshire Council Agr. Educ. (Pamphlet), no. 75, p. 36.

(50) CROWTHER, C., AND RUSTON, A. G. 1915 The influence on crop and soil of manures applied to permanent meadow. *In* Jour. Agr. Sci., v. 7, no. 2, p. 197–218.

(51) D'ALBUQUERQUE, J. P., AND BOVELL, J. R. 1910 Variety and manurial experiments with sugar cane. Rpt. Agr. Work Imp. Dept. Agr. West Indies, 1908–1910, p. 4–109.

(52) DAMSEAUX, A. 1910 Sea salt in sugar beet production. *In* Jour. Soc. Agr. Brabant et Hainaut, t. 55, no. 48, p. 1175–1176.

(53) DANNFELT, H. J., AND RHODIN, S. 1906 Culture tests with potatoes in 1905. *In* K. Landtbr. Akad. Handl. och Tidskr., v. 45, no. 2, p. 92–131.

(54) DENEUMOSTIER, C. 1906 The relation of temperature and humidity to the germination of seed of certain grasses. *In* Bul Agr. (Brussels), t. 22, no. 7, p. 983–998.

(55) DUGGAR, J. F. AND CAUTHEN, E. F. 1913 Oats, experiments on culture, varieties and fertilization. Ala. Agr. Exp. Sta. Bul. 173, p. 121–144.

(56) EASTERBY, H. T. 1914 Fertilizers on sugar cane. *In* Ann. Rpt. Bur. Sugar Exp. Sta. (Queensland), 1914, p. 30, 31.

(57) ETHERIDGE, W. C. 1911 Report of division of agronomy. *In* N. C. Agr. Exp. Sta. Ann. Rpt., p. 16–21.

(58) EWART, A. J. 1914 The influence of salt on plants. *In* Jour. Dept. Agr. Victoria, v. 12, no. 7, p. 420–423.

(59) VON FEILITZEN, H. 1909 The effect of lime nitrogen containing a high percentage of calcium carbid on plant growth. *In* Deut. Landw. Presse, Bd. 36, No. 30, p. 327.

(60) VON FEILITZEN, H. 1915 Culture experiments on moor soils at Flahult and Torestorp, Sweden. *In* Svenska Mosskulturför. Tidskr., v. 29, no. 6, p. 441–473; v. 30, no. 1, p. 51–72; no. 2, p. 119–159.

(61) FERUGLIO, D. 1906 Nitrate of soda as compared with a mixture of nitrate of soda and sulfate of ammonia. *In* Ann. R. Staz. Sper. Agr. Udine, v. 8, p. 72–76.

(62) FRED, E. B. 1916 Relation of green manuring to the failure of certain seedlings. *In* Jour. Agr. Res. v. 5, no. 25, p. 1161–1176.

(63) FRED, E. B. 1916 Work with field crops in 1915. Wis. Agr. Exp. Sta. Bul. 268, p. 4–6; 10–15; 32, 36.

(64) GARDNER, F. D., AND BROWN, B. E. 1911 Growth of clover as influenced by lime in different forms and amounts when applied to soil from the general fertilizer plats. *In* Penn. Agr. Exp. Sta. Ann. Rpt., p. 60–76.

(65) GARDNER, F. D., et al 1917 Thirty-five years' results with fertilizers. *In* Penn. Agr. Exp. Sta. Bul. 146, p. 3–29.

(66) GASKILL, E. F. 1914 Fertilizer experiments. *In* Mass. Agr. Sta. Exp. Ann. Rpt., p. 30a–32a.

(67) GAUL, F. 1912 Fertilizer tests of oats. In Illus. Landw. Ztg., Bd. 32, No. 10, p. 75, 76.

(68) GAUL, F. 1913 The influence of fertilizing on the resistance of grain to hail. *In* Deut. Landw. Presse, Bd. 40, No. 104, p. 1252.

(69) GERLACH, M., et al. 1914 Field experiments on the action of different nitrogenous fertilizers. *In* Ber. Landw. Reichsamte Innern, No. 34, p. 229.

(70) GILCHRIST, D. A. 1909 Work with field crops. County Northumb., Ed. Com., Bul. 13, p. 42–63.

(71) GILCHRIST, D. A. 1909 Experiments with field crops. County Northumb, Ed. Com., Bul. 14, p. 38–66.

(72) GILCHRIST, D. A. 1911 Guide to experiments for 1911. County Northumb., Ed. Com., Bul. 16, p. 5–73.

(73) GOESSMANN, C. A. 1891 Field experiments. *In* Mass. Agr. Exp. Sta. Ann. Rpt. 1891, p. 149–158.

(74) GOFF, E. S. 1897 The influence of varying amounts of water on the germination of beet seeds. *In* Wis. Agr. Exp. Sta. Ann. Rpt. 1897, p. 299–302.

(75) GRAHL, H. 1895 Comparative tests during 1893 and 1894 of sulfate of ammonia and nitrate of soda. *In* Jahrb. Deut. Landw. Gesell., Bd. 10, p. 482–487.

(76) GRANDEAU, L. 1903 Nitrate of soda and sulfate of ammonia. *In* Jour. Agr. Prat., n. s., t. 5, no. 10, p. 308–309.

(77) GRANDEAU, L. 1906 Comparative tests of the fertilizing value of nitrate of soda, sulfate of ammonia and calcium cyanamid. *In* Jour. Agr. Prat., n. s., t. 11, no. 1, p. 8–10.

(78) DE GRAZIA, S. 1909 On the efficiency of soluble fertilizer salts in dry climates. *In* Ann. R. Staz. Agr. Sper. Roma, ser. 2, v. 3, p. 157–186.

(79) DE GRAZIA, S., AND CALDIERI, S. 1906 The influence of fertilizer applications on the composition of the grain of rye. *In* Staz. Sper. Agr. Ital., v. 39, no. 6–7, p. 514–528.

(80) GREIG, R. B. 1909 Report on turnips manuring experiment, 1909. Aberdeen and No. Scot. Col. Agr. Leaflet 11, p. 6.

(81) HALL, A. D. 1909 Some secondary actions of manures upon the soil. *In* Rothamsted Mem., v. 8, 1902–1912, p. 1–24; also *in* Jour. Roy. Agr. Soc., v. 70, p. 12–35.

(82) HALL, A. D. 1910 The new nitrogenous fertilizers, calcium cyanamid, and nitrate of lime. *In* Jour. Bd. Agr. (London), v. 16, no. 12, p. 1006–1008.

(83) HAMPE, W. 1867 Vegetations-Versuche mit Ammoniaksalzen, Hurnsaure, Hippursaure und Glycocol, als stickstoffhaltigen Nahrungs-mitteln der Pflanzen. *In* Landw. Vers. Stat., Bd. 10, p. 175.

(84) HANSEN, F. 1910 Experiments with different nitrogenous fertilizers for spring grains and root crops. *In* Tidskr. Landbr. Planteavl, v. 17, no. 5, p. 693–731.

(85) HARPER, J. N. 1908 Fertilizer experiments with cotton. S. C. Agr. Exp. Sta. Bul. 145, p. 3–31.

(86) HARRIS, J. E., AND GORTNER, R. A. 1914 Notes on the calculation of the osmotic pressure of expressed vegetable saps from the depression of the freezing point, with table for values of P from 0.001° to 2.999°. *In* Amer. Jour. Bot., v. 1, p. 75–78.

(87) HARRIS, F. S. 1914 Effect of variation in moisture content on certain properties of a soil and on the growth of wheat. N. Y. (Cornell) Agr. Exp. Sta. Bul. 352, p. 805–868.

(88) HARRISON, J. B. 1908 Effect of soluble salts in the soil on the growth of sugar cane. *In* West Indian Bul., v. 9, no. 1, p. 19–37.

(89) HARRISON, J. B. 1909 Variety and manurial tests with cane, rice and cotton. *In* Rpt. Exp. Agr. Work Dept. Sci. and Agr. (Brit. Guiana), 1908–9, p. 1–15, 21–27.

(90) HARRISON, J. B. et al. 1914 The crops on the experimental sugar cane fields. *In* Jour. Bd. Agr. Brit. Guiana, v. 8, no. 1–2, p. 45–57.

(91) HASELHOFF, E. 1914 Experiments with nitrogenous fertilizers. *In* Landw. Vers. Stat., Bd. 84, No. 1–2, p. 1–55.

(92) HASELHOFF, E. 1915 The action of chlorides on soil and plant. *In* Fuhling's Landw. Ztg., Bd. 64, No. 19–20, p. 478–508.

(93) HEINE 1911 Fertilizer experiments with small fruits, vegetables and flowers. *In* Ber. K. Gart. Lehranst. Dahlem, 1908 and 1909, p. 195–204.

(94) HELLRIEGEL, H. 1861 *In* Ann. Landw., v. 7, p. 53; v. 8, p. 119; cited by S. W. Johnson in How Crops Feed, Orange Judd Co., N. Y. (1883), p. 302.

(95) HELLRIEGEL, H. 1883 Beiträge zur den naturwissenschaftlichen Grundlagen des Ackerbaus, u.s.w. Cited by Russell, E. J., Soil Conditions and Plant Growth (1915), London, p. 27.

(96) HELLRIEGEL, H. 1898 Vegetationsversuche über den Kilibedarf einiger Pflanzen. *In* Arb. Deut. Landw. Gesell., No. 34.

(97) HENDRICK, J. 1908 Experiments with nitrogenous manures 1904–1908. *In* Aberdeen and No. Scot. Col. Agr. Bul. 13, p. 29.

(98) HERKE, S. 1915 The action of the nitrogen of sodium nitrate, ammonium sulfate and lime nitrogen. *In* Kiserlet Közlem., Bd. 18, No. 2, p. 266–306.

(99) HICKS, G. H. 1898 The effect of fertilizers on the germination of seeds. *In* Proc. Amer. Assoc. Adv. Sci., v. 47, p. 428–429.

(100) HILTNER, L. 1915 The effect of different nitrogenous fertilizers, especially guanidin nitrate, on oats and mustard. *In* Prakt. Bl. Pflanzenbau u. Schutz., n. s., Bd. 13, No. 11-12, p. 141–155.

(101) HILTNER, L., AND LANG, F. 1914 Experiments on the action of different air-nitrogen fertilizers. *In* Prakt. Bl. Pflanzenbau u. Schutz., n. s., Bd. 12, No. 11, p. 121–128.

(102) HOFFMANN, C. 1913 Soil and fertilizer investigations. Wis. Agr. Exp. Sta. Bul. 228, p. 26, 27.

(103) HUNT, T. F. 1909 Soil fertility. Penn. Agr. Exp. Sta. Bul. 90, p. 26.

(104) HUNT, T. F., et al 1912 Summary of the results obtained from experiments with commercial fertilizers, yard manure, lime, etc., extending through a period of thirty years. *In* Penn. Agr. Exp. Sta. Ann. Rpt. 1912, p. 83–119.

(105) HUNTER, C. 1912 Some observations on the effect of soil aeration on plant growth. *In* Proc. Univ. Durham Phil. Soc., v. 4, no. 4, p. 183–186.

(106) HUSTON, H. A. 1892 Forms of nitrogen for wheat. Ind. Agr. Exp. Sta. Bul. 41, p. 100–102.

(107) HUTCHISON, H. B., AND MILLER, N. H. J. 1909 Direct assimilation of ammonium salts by plants. *In* Jour. Agr. Sci., v. 3, p. 179–94.

(108) JACOMETTI, G. 1908 The new nitrogenous fertilizers in comparison with the old. *In* Ann. Accad. Agr. Torino, v. 51, p. 179–208.

(109) DE JONG, A. W. K. 1910 Field experiments with fertilizers. *In* Teysmannia, v. 21, no. 4–5, p. 291–301.

(110) DE JONG, A. W. K. 1912 Fertilizer experiments with field crops. *In* Dept. Landb. Nijv. en Handel (Dutch East Indies), Meded. Agr. Chem. Lab., no. 1, p. 40.

(111) DE JONG, A. W. K. 1915 Fertilizer experiments. Dept. Landb. Nijv. en Handel (Dutch East Indies), Meded. Agr. Chem. Lab., no. 13, p. 27.

(112) KALINKIN, S. I. 1912 The utilization of ammonical nitrogen by corn plantlets. Izv. Moskov. Selsk. Khoz. Inst. (Ann. Inst. Agron. Moscou), v. 18, no. 1, p. 180–192.

(113) KAPPEN, H. 1915 Fertilizer experiments with transformation products of lime nitrogen. *In* Land Vers. Stat., Bd. 86, No. 1–2, p. 115–136.

(114) KELKAR, G. K. 1914 Manurial experiments. Dept. Agr. Bombay, Ann. Rpt. Expt. Work Surat Agr. Sta., p. 23–31.

(115) KELLEY, W. P. 1912 The assimilation of nitrogen by rice. Hawaii Agr. Exp. Sta. Bul. 24, p. 2.

(116) KELLEY, W. P., et al. 1911 Field crops at the Hawaii Station. *In* Hawaii Agr. Exp. Sta. Ann. Rpt. 1911, p. 9, 51–63.

(117) KIMBROUGH, J. M. 1912 Cotton culture. Ga. Agr. Exp. Sta. Bul. 100, p. 229–248.

(118) KIRCHNER 1901 Sulfate of ammonia as a top dressing for winter wheat. *In* Deut. Landw. Presse, Bd. 28, No. 21, p. 171.

(119) KLEBERGER 1907 Nitrate of soda and ammonium sulfate for cereals. *In* Deut. Landw. Presse, Bd. 34, No. 15, p. 119–120.

(120) KLOEPFER, G. 1899 Top dressing of sulfate of ammonia and nitrate of soda. *In* Fuhling's Landw. Ztg., Bd. 48, No. 3, p. 114–116; No. 4, p. 138–142.

(121) KLOEPFER, G. 1900 Results of fertilizer experiments with sulfate of ammonia. *In* Fuhling's Landw. Ztg., Bd. 49, No. 10, p. 376–384; No. 11, p. 396–406; No. 12, p. 436–445.

(122) KLOEPFER, G. 1902 Field experiments with ammonium sulfate in the province of Posen. *In* Fuhling's Landw. Ztg., Bd. 51, No. 6, p. 193–197; No. 7, p. 231–233.

(123) KRAUSS, F. G. 1900 Fertilizer experiments with sulfate of ammonia and nitrate of soda. *In* Fuhling's Landw. Ztg., Bd. 49, No. 6, p. 232–237; No. 7, p. 256–259.

(124) KRAUSS, F. G. 1907 Rice investigations; Report of first year's experiment. *In* Hawaii Agr. Exp. Sta. Ann. Rpt. 1907, p. 67–90.

(125) KRAUSS, F. G. 1908 Field crop experiments. *In* Hawaii Agr. Exp. Sta. Ann. Rpt. 1908, p. 65–84.

(126) KRETSCHMER, et al 1906 Field experiments on the fertilizing action of sulfate of ammonia as compared with nitrate of soda. *In* Arb. Deut. Landw. Gesell., No. 121, p. 23–234.

(127) LARSEN, O. H. 1912 Reports on cooperative fertilizer and variety tests, Zeeland, 1912. Ber. Landbofor. Virks. Planteavl. Sjælland, 1912, p. 450.

(128) LAWES, J. B., GILBERT, J. H., AND PUGH, E. 1861 On the sources of the nitrogen of vegetation. *In* Phil. Trans. Roy. Soc. London, pt. 2, p. 431–587.

(129) LAWES, J. B., AND GILBERT, J. H.; cited by A. D. Hall in The Book on the Rothamsted Experiments, New York (1905), p. 31–239.

(130) LILIENTHAL. 1904 Sulfate of ammonia or nitrate of soda? *In* Fuhling's Landw. Ztg., Bd. 53, No. 4, p. 129–134.

(131) LILIENTHAL 1906 Is there any considerable evaporation of ammonia from soils? *In* Illus. Landw. Ztg., Bd. 26, No. 99, p. 851–852.

(132) LIPMAN, C. B., AND GERICKE, W. F. 1916 A vegetation experiment on the availability of nitrogenous fertilizers in an arid soil. *In* Soil Sci., v. 2, p. 575–581.

(133) LIPMAN, J. G. 1915 Factors relating to the availability of nitrogenous plant foods. N. J. Agr. Exp. Sta. Bul. 251, p. 3–55.

(134) LIPMAN, J. G., AND BLAIR, A. W. 1915 The availability of nitrogenous materials. *In* N. J. Agr. Exp. Sta. 36th Ann. Rpt., p. 195–203.

(135) LIPMAN, J. G., AND BLAIR, A. W. 1916 Investigations relative to the use of nitrogenous plant foods, 1898–1912. N. J. Agr. Exp. Sta. Bul. 288, p. 126.

(136) LIPMAN, J. G., et al 1912 Conditions affecting the availability of nitrogen compounds in vegetation experiments. *In* N. J. Agr. Exp. Sta. 33rd Ann. Rpt., p. 205–233.

(137) LIPMAN, J. G., et al 1914 Pot experiments on the availability of nitrogen in mineral and organic compounds. *In* N. J. Agr. Exp. Sta. 35th Ann. Rpt., p. 207–220.

(138) LIPMAN, J. G., et al 1912 Conditions affecting the availability of nitrogenous compounds in vegetation experiments. N. J. Agr. Exp. Sta. Bul. 249, p. 3–23.

(139) LIVINGSTON, B. E. 1907 The relation of desert plants to soil moisture and to evaporation. *In* Carnegie Inst. Washington Pub., v. 50, p. 78.

(140) MALPEAUX, L. 1911 The use of cyanamid as a fertilizer. *In* Jour. Agr. Prat., n. s., t. 22, no. 47, p. 647–651.

(141) MALPEAUX, L. 1913 Nitrogenous fertilizers for field crops. *In* Jour. Agr. Prat., n. s., v. 25, no. 7, p. 237–238.

(142) DE MARNEFFE, G. 1896 On the influence of fertilizers on germination. *In* Ingen. Agr. Gembloux, ann. 6, no. 11, p. 490–491.

(143) MAUSBERG, A. 1913 The influence of fertilization on the conditions of the soil and its fitness for certain crops. *In* Landw. Jahrb., Bd. 45, No. 1, p. 29–101.

(144) MAUSBERG, A. 1914 The effect of sodium nitrate and ammonium sulfate on potatoes and sugar beets. *In* Landw. Jahrb., Bd. 46, No. 2, p. 339–342.

(145) MAYER, A. 1898 Ueber den Einfluss kleiner oder grossere Mengen von Wasser auf die Entwickelung einiger Kulturpflanzen. *In* Jour. Landw., Bd. 46, p. 167–184.

(146) MAYER, A. 1909 Nitrate of soda or sulfate of ammonia. *In* Agr. Jour. Cape Good Hope, v. 35, no. 1, p. 24–27.

(147) MCCALL, A. G. 1916 Physiological balance of nutrient solutions for plants in sand cultures. *In* Soil Sci., v. 2, no. 3, p. 207–253.

(148) MCCLELLAND, C. K. 1916 Report of field crops work at the Georgia Experiment Station. *In* Ga. Agr. Exp. Sta. Ann. Rpt. 1916, p. 7–9, 28–31.

(149) MCCOOL, M. M., AND WHEETING, L. C. 1917 Movement of soluble salts through soils. *In* Jour. Agr. Res., v. 2, p. 531–547.

(150) MILBURN, T. 1908 Report on field trials on the manuring of potatoes. Midland Agr. and Dairy Col. Bul. 5, p. 43–50.

(151) MILBURN, T., AND GAUT, R. C. 1911 Report of experiments with nitrogenous manures. County Council Lancaster, Ed. Com., Agr. Dept., Farmer's Bul. 20, p. 10.

(152) MITSCHERLICH, E. A. 1912 Das Wasser als Vegetations-factor. *In* Landw. Jahrb., Bd. 42, p. 701–717.

(153) MORGAN, J. O. 1911 The effect of soil moisture and temperature on the availability of plant nutrients in the soil. *In* Proc. Amer. Soc. Agron., v. 3, p. 191–249.

(154) MOROSOV, V. A. 1914 The role of calcium carbonate in the assimilation of ammonia. *In* Iz Result. Veget. Opytov Lab. Rabot, Izv. Moscov. Selsk. Khoz. Inst. (Ann. Inst. Agron. Moscou), v. 10, p. 390–395.

(155) NAZARI, V. 1908 The comparative fertilizing value of calcium cyanamid, calcium nitrate, ammonium sulfate and nitrate of soda. *In* Atti R. Accad. Lincei, Rend. Cl. Sci. Fis., Mat. e Nat., ser. 5, v. 17, no. 7 p. 334–342.

(156) NIKLEVSKI, B. 1915 The influence of the distribution of nitrogenous fertilizers and straw in soil on plant production. *In* Ztschr. Landw. Versuchsw. Osterr., Bd. 18, No. 12, p. 674–90.

(157) NIKOLAEVA, A. G. 1914 The formation of asparagin by *Lupinus luteus*. *In* Iz Result. Veget. Opytov Lab. Rabot, Izv. Moscov. Selsk. Khoz. Inst. (Ann. Inst. Agron. Moscow), v. 10, sup. 3, p. 280–283.

(158) ORCHINNIKOV, N. 1910 The relative effect of different forms of nitrogen on the yield of oats in dependence upon the character of the soil and the conditions of the experiments. *In* Zhur. Opuitn. Agron. (Russ. Jour. Expt. Landw.), v. 11, no. 4, p. 481–532.

(159) OSWALD, S., et al 1914 Observations on the relative value of the most important nitrogen fertilizers. *In* Landw. Jahrb., Bd. 47, No. 1, p. 79–106.

(160) OTTO, R. 1904 Comparative fertilizer and vegetative experiments with lime nitrogen on garden vegetables. *In* Gartenflora, Bd. 53, No. 20, p. 534–538.

(161) OTTO, R. 1908 Comparative fertilizer experiments with nitrogen lime, lime nitrogen, nitrate of soda, and sulfate of ammonia for sugar beets. *In* Deut. Landw. Presse, Bd. 35, No. 1, p. 1.

(162) PANTANELLI, E., AND SEVERINI, G. 1910 Some experiments on the utilization of

ammonium salts by green plants. *In* Staz. Sper. Agr. Ital., v. 43, no. 6, p. 449–544.

(163) PANTANELLI, E., AND SEVERINI, G. 1911 Further experiments on the utilization of ammonium salts by green plants. *In* Staz. Sper. Agr. Ital., v. 44, no. 11–12, p. 873–908.

(164) PARSONS, T. S. 1911 Fertilizer and variety tests with field crops. *In* Wyo. Agr. Exp. Sta. Ann. Rpt. 1911, p. 41–45.

(165) PATTERSON, H. J. 1904 Experiments with nitrogenous fertilizers. Md. Agr. Exp. Sta. Bul. 94, p. 25–47.

(166) PERITURIN, F. T. 1912 Nutritive changes in sprouting pumpkin seeds. *In* Izv. Moskov. Selsk. Khoz. Inst. (Ann. Inst. Agron. Moscou), v. 18, no. 2, p. 228–256.

(167) PETROV, G. G. 1911 Nitrogen assimilation under sterile conditions of plants from nitrates, ammonium salts, and asparagin. *In* Izv. Moskov. Selsk. Khoz. Inst. (Ann. Inst. Agron. Moscou), v. 17, no. 4, p. 141–178.

(168) PITSCH, O. 1887 Versuche zur Entscheidung der Frage, ob salpetersaure Salze für die Entwicklung unserer landwirtschaftlichen Kilturgewächse unenteberlich sind oder nicht. *In* Landw. Vers. Stat., Bd. 34, p. 217–258.

(169) POPP, M. 1916 Pot fertilizer experiments with new nitrogenous fertilizer. *In* Mitt. Deut. Landw. Gesell., Bd. 31, No. 4, p. 54–57.

(170) PORTER, E., AND GAUT, R. C. 1907 Report of an experiment on the manuring of rye grass and clover at several centers in the country during 1906–7. County Council Lancaster, Ed. Com., Agr. Dept., Farmer's Bul. 15, p. 23.

(171) PORTER, J., AND MCWILLIAM, A. S. 1915 Report on the manuring of mangels. Herefordshire Ed. Com., Farmer's Bul. 9, p. 9.

(172) POSPISIL, K. 1914 Sodum nitrate vs. ammonium sulfate. *In* Wiener Landw. Ztg., Bd. 64, No. 18–19, p. 163–164.

(173) PRIANISHNIKOV, D. 1900 The influence of the moisture of the soil on the growth of plants. *In* Zhur. Opuitn. Agron. (Russ. Jour. Expt. Landw.), v. 1, no. 1, p. 3–20.

(174) PRIANISHNIKOV, D. N. 1913 Concerning the relation of etiolated shoots of maize and lupine to ammonium salts and nitrates. *In* Iz Result. Veget. Opytov Lab. Rabot, Izv. Moscov. Selsk. Khoz. Inst. (Ann. Inst. Agron. Moscow), v. 9, p. 559–565.

(175) PRIANISHNIKOV, D. N. 1913 The synthesis of amids which depend on ammonia absorbed through roots. *In* Rev. Gén. Bot., t. 25, no. 289, p. 5–13.

(176) PRIANISHNIKOV, D. N. 1914 The role of ammonia in the transformation of nitrogenous material in plants. *In* Iz Result. Veget. Opytov. Lab. Rabot, Izv. Moscov. Selsk. Khoz. Inst. (Ann. Inst. Agron. Moscow), v. 10, sup. 3, p. 24.

(176a) PROMSY, G. 1911 The influence of acidity on germination. *In* Compt. Rend. Acad. Sci. (Paris), t. 152, no. 8, p. 450–452.

(176b) PROMSY, G. 1912 The role of acids in germination. *In* Bul. Soc. Nat. Agr. France, t. 72, no. 10, p. 916–922.

(177) RAWSON, W. B., et al. 1909 Experiments with field crops. *In* New Zeal. Dept. Agr. Ann. Rpt., v. 17, p. 123–132; 350–358; 365–373; 385–434; 442–447.

(178) RINDELL, A. 1914 Experiments with nitrogenous fertilizers at the Finnish moor culture Experiment Station in 1911 to 1913. *In* Finska. Mosskulturfor. Arsbok, v. 18, no. 1, p. 53–98.

(179) RITMAN, G. I. 1912 The formation of asparagin in sprouting vetches. *In* Izv. Moskov. Selsk. Khoz. Inst. (Ann. Inst. Agron. Moscou), v. 18, no. 2, p. 212–220.

(180) RHODIN, S. 1891 Fertilizer experiments with sulfate of ammonia and nitrate of soda for barley and oats. *In* K. Landtbr. Akad. Handl. och Tidskr., v. 30, p. 142–148; v. 32, p. 78–82.

(181) RHODIN, S. 1907 Comparative fertilizer trials with new nitrogenous fertilizers. *In* K. Landtbr. Akad. Handl. och Tidskr., v. 46, no. 1, p. 3–21.

(182) RHODIN, S. 1912 Experiments with new nitrogenous fertilizers. *In* K. Landtbr. Akad. Handl. och Tidskr., v. 51, no. 5, p. 362–382.

(183) RHODIN, S. 1913 Nine years experiments with the new nitrogenous fertilizers. *In* Meddel. Centralanst. Försöksv. Jordbruksomradet, no. 75, p. 17.

(184) RUPRECHT, R. W., AND MORSE, F. W. 1915 The effect of sulfate of ammonia on soil. Mass. Agr. Exp. Sta. Bul. 165, p. 73–90.

(185) RUSCHE, A. 1912 Influence of various fertilizer salts on the germination of field crop seeds. *In* Jour. Landw., Bd. 60, No. 4, p. 305–365.

(186) RUSSELL, E. J. 1915 Soil Conditions and Plant Growth, p. 49. London.

(187) SACHS, J., AND KNOP, W. Cited by Johnson, S. W., How Crops Feed, (1883), New York, p. 302.

(188) SCHMITZ, N. 1917 Timothy fertilization and culture. Md. Agr. Exp. Sta. Bul. 202, p. 163.

(189) SCHMOEGER, M. 1907 Comparative fertilizer experiments with lime nitrogen. *In* Mitt. Deut. Landw. Gesell., Bd. 22, No. 10, p. 103–105.

(190) SCHNEIDEWIND, W. 1907 Experiments with nitrogenous fertilizers at the Halle Experiment Station during 1905–1906. *In* Landw. Wchnschr. Sachsen, Bd. 9, No. 9, p. 87–89.

(191) SCHNEIDEWIND, W., et al 1907 Fertilizer experiments. *In* Landw. Jahrb., Bd. 36, No. 4, p. 586–608.

(192) SCHNEIDEWIND, W., et al 1908 Experiments on the action of nitrate of soda, ammonium sulfate, lime nitrogen, nitrogen lime and the Norwegian lime niter. *In* Arb. Deut. Landw. Gesell., No. 146, p. 118.

(193) SCHNEIDEWIND, W., et al 1910 Experiments with nitrogenous fertilizers. *In* Landw. Jahrb., Bd. 39, Erganzungsb. 3, p. 209–236.

(194) SCHNEIDEWIND, W. et al 1910 Fertilizer experiments. *In* Landw. Jahrb., Bd. 39, Erganzungsb. 3, p. 20–109.

(195) SCHNEIDEWIND, W., et al 1912 Further experiments on the action of various forms of nitrogen. *In* Arb. Deut. Landw. Gesell., No. 217, p. 171.

(196) SCHNEIDEWIND, W., et al 1916 Field experiments on the action of new forms of nitrogen. *In* Mitt. Deut. Landw. Gesell., Bd. 31, No. 2, p. 16–20.

(197) SCHREIBER, C. 1908 Experiments with nitrogenous fertilizers. *In* Rev. Gén. Agron., n. s., t. 3, no. 3, p. 97–105.

(198) SCHULZE, B. 1907 Experiments with lime nitrogen as a fertilizer. *In* Fuhling's Landw. Ztg., Bd. 56, No. 5, p. 145–159.

(199) SCHULZE, B. 1910 The fertilizer action of salt in combination with ammonium sulfate. *In* Mitt. Deut. Landw. Gesell., Bd. 25, No. 30, p. 452–458.

(200) SCHULZE, B. 1913 Beitrage zur Frage der Dungung mit Natronsalzen. *In* Landw. Vers. Stat., Bd. 79–80, p. 431.

(201) SEBELIEN, J. 1907 Further experiments with various kinds of nitrate. *In* Jour. Landw., Bd. 55, No. 4, p. 293–297.

(202) VON SEELHORST, C. 1899 Ueber den Wasserverbrauch der Haferpflanze bei verschiedenem Wassergehalt und bei verschiedener Dungung des Bodens. *In* Jour. Landw., Bd. 47, p. 369–378.

(203) VON SEELHORST, C. 1911 The importance of water in the life of cultivated plants. *In* Jour. Landw., Bd. 59, No. 3, p. 259–291.

(204) VON SEELHORST, C., AND TUCKER, M. 1898 Der Einfluss, welcher der Wasserhalt und der Reichtum des Bodens auf die Ausbildung der Wurzeln und der oberirdischen Organe der Haferpflanzen ausuben. *In* Jour. Landw., Bd. 46, p. 52–63.

(205) VON SEELHORST, C., et al 1900 Einfluss des Wassergehaltes und der Dungung des Bodens auf die Produktion und die Zusamenstzung von Futterpflanzen, italienisches Raingrass und Klee. *In* Jour. Landw., Bd. 48, p. 265–86.

(206) SHIVE, J. W. 1915 A study of physiological balance in nutrient media. *In* Physiol. Res., v. 1, No. 7, p. 327–397.

(207) SHULOV, I. 1912 Studies of sterile cultures of maize in regard to nitrogen assimilation. *In* Zhur. Opuitn. Agron. (Russ. Jour. Expt. Landw.), v. 13, no. 2, p. 200–206.

(208) SOAVE, M. 1908 The action of gypsum as a fertilizer. *In* Staz. Sper. Agr. Ital., v. 41, no. 8, p. 473–500.

(209) SODERBAUM, H. G. 1908 Results of trials with nitrogenous fertilizers. *In* K. Landtbr. Akad. Handl. och Tidskr., v. 47, no. 1–2, p. 104–110.

(210) SODERBAUM, H. G. 1908 The factors which influence the fertilizing effect of difficultly soluble phosphate. *In* Landw. Vers. Stat., Bd. 68, no. 5–6, p. 433–450.

(211) SODERBAUM, H. G. 1910 Ammonium sulfate as a fertilizer. *In* Meddel. Centralanst. Försöksv. Jordbruksomradet, no. 26, p. 17.

(212) SODERBAUM, H. G. 1915 Certain factors which influence the fertilizing action of the slightly soluble phosphates. *In* K. Landtbr. Akad. Handl. och Tidskr., v. 54, no. 5, p. 448–477.

(213) STEBUTT, A. 1907 On the influence of certain lime compounds on the fertilizing value of ammonium sulfate and nitrogen lime. *In* Fuhling's Landw. Ztg., Bd. 56, no. 19, p. 669–676.

(214) STEGLICH, 1907 On fertilizing experiments with lime nitrogen, nitrogen lime and lime niter. *In* Fuhling's Landw. Ztg., Bd. 56, No. 22, p. 769–781.

(215) STOKES, E. E. 1911 Results of field trials on the manuring of sweedes in the year 1910. Midland Agr. and Dairy Col. Bul. 7, p. 43–47.

(216) STUTZER, A. 1907 Investigations on the action of heavy applications of ammonium sulfate in presence of organic substances and calcium carbonate in the soil. *In* Jour. Landw., Bd. 55, No. 1, p. 81–91.

(217) STUTZER, A. 1908 Fertilizer experiments with nitrogen lime for potatoes. *In* Jour. Landw., Bd. 56, No. 2, p. 141–144.

(218) SÜCHTING, H. 1907 Culture experiments and critical studies on the relative efficiency of nitrate of soda and sulfate of ammonia. *In* Jour. Landw., Bd. 55, No. 1, p. 1–46.

(219) SVOBODA, H. 1910 A comparison of nitrate of soda and ammonium sulfate as fertilizers for oats. *In* Ztschr. Landw. Versuchsw. Osterr., Bd. 13, No. 10, p. 812–828.

(220) SWANSWICK, B., AND KINCH, E. 1909 Field and manurial experiments with potatoes and mangels. *In* Agr. Students' Gaz., n. s., v. 14, no. 5, p. 162–167.

(221) SWANSWICK, B., AND KINCH, E. 1910 Manurial experiments on oats at Coates. *In* Agr. Students' Gaz., n. s., v. 15, no. 2, p. 43, 44.

(222) TACKE, B., AND BRUNE, F. 1913 Comparative tests of lime nitrogen, nitrogen lime, sodium nitrate and ammonium sulfate on sandy and upland moor soils. *In* Landw. Vers. Stat., Bd. 83, No. 1–2, p. 1–100.

(223) TAKEUCHI, T. 1908 Gypsum as a manure. *In* Bul. Col. Agr., Tokyo Imp. Univ., v. 7, no. 5, p. 583–597.

(224) TEMPANY, H. A. 1910 Manurial experiments with cotton in the Leeward Islands. West Indian Bul., v. 11, no. 1, p. 60–67.

(225) TOLF, R. 1898 On the influence of humus soils on germination. *In* Tidskr. Landtmän, v. 19, no. 22, p. 387–390.

(226) TOTTINGHAM, W. E. 1914 A quantitative chemical and physiological study of nutrient solution for plant cultures. *In* Physiol. Res., v. 1, p. 133–245.

(227) TOWAR, J. D. 1892 Cooperative field experiments with fertilizers on corn. *In* R. I. Agr. Exp. Sta. Ann. Rpt. 1892, p. 163–198.

(228) TULAIKOFF, N. 1915 Transpiration coefficient of cultural plants. *In* Zhur. Opuitn. Agron. (Russ. Jour. Expt. Landw.), v. 16, no. 1, p. 36.

(229) VANHA, J. J., et al. 1909 The action of lime nitrogen, nitrate of soda and sulfate of ammonia. *In* Ztschr. Landw. Versuchsw. Osterr., Bd. 12, No. 12, p. 785–838.

(230) VAN HOECK, P., AND RAUWERDA, A. 1911 Fertilizer experiments. *In* Dept. Landb., Nijv. en Handel, Verslag. en Meded.Dir. Landb. (Netherlands), no. 1, p. 1–44.

(231) VILLE, M. G. 1857 Recherches experimentales sur la vegetation. Also cited by Lawes, J. B., Gilbert, J. H., and Pugh, E. in "On the sources of the nitrogen of vegetation." *In* Philos. Trans., 1861, pt. 2, p. 431–577.

(232) VIVIER, A. 1896 Comparative trial of nitrate of soda and sulfate of ammonia with barnyard manure for sugar beets. *In* Ann. Sci. Agron., ser. 2, v. 2, I, no. 3, p. 384–386.

(233) VOELCKER, J. A. 1906 The Woburn field experiments, 1905–1906. *In* Jour. Roy. Agr. Soc. England, v. 67, p. 282–310.

(234) VOELCKER, J. A. 1907 The Woburn field experiments. *In* Jour. Roy. Agr. Soc. England, v. 68, p. 242–266.

(235) VOELCKER, J. A. 1908 The Woburn field experiments. *In* Jour. Roy. Agr. Soc. England, v. 69, p. 337–357.

(236) VOELCKER, J. A. 1908 Pot culture experiments. *In* Jour. Roy. Agr. Soc. England, v. 70, p. 388–395.

(237) VOELCKER, J. A. 1909 The Woburn field experiments. *In* Jour. Roy. Agr. Soc. England, v. 70, p. 362–388.

(238) VOELCKER, J. A. 1911 The Woburn field experiments. *In* Jour. Roy. Agr. Soc. England, v. 72, p. 387–403.

(239) VOELCKER, J. A. 1914 The Woburn field experiments. *In* Jour. Roy. Agr. Soc. England, v. 75, p. 286–306.

(240) VOELCKER, J. A. 1915 The Woburn field experiments. *In* Woburn Exp. Sta. Rpt., p. 1–15; 20–29; also *in* Jour. Roy. Agr. Soc. England, v. 76, p. 317–330; 334–344.

(241) VOELCKER, J. A. 1916 Ammonium sulfate on continuous fields. *In* Jour. Roy. Agr. Soc. England, v. 77, p. 235–64.

(242) VOORHEES, E. B. 1901 Investigations relative to the use of nitrogenous materials. *In* N. J. Agr. Exp. Sta. 22nd Ann. Rpt., p. 144–184.

(243) VOORHEES, E. B. 1903 Investigations relative to the use of nitrogenous materials. *In* N. J. Agr. Exp. Sta. 24th Ann. Rpt., p. 148–184.

(244) VOORHEES, E. B. 1904 Investigations relative to the use of nitrogenous materials. *In* N. J. Agr. Exp. Sta. 25th Ann. Rpt., p. 191–233.

(245) VOORHEES, E. B., AND LIPMAN, J. G. 1906 Investigations relative to the use of nitrogenous materials. *In* N. J. Agr. Exp. Sta. 27th Ann. Rpt., p. 71–100.

(246) VOORHEES, E. B., AND LIPMAN, J. G. 1909 Investigations relative to the use of nitrogenous materials. N. J. Agr. Exp. Sta. Bul. 221, p. 3–52.

(247) WAGNER, P. 1868 Vegetationversuche über die Stickstoff-Ernahrung der Pflanzen. *In* Landw. Vers. Stat., Bd. 11, p. 287.

(248) WAGNER, P. 1881 Beitrage zur Bergrundung und Ausbildung einer exakten Methode der Dungungsversuche. *In* Jour. Landw., Bd. 28, p. 9.

(249) WAGNER, P. 1901 Experiments on the relative fertilizing value of ammonium salts. *In* Mitt. Deut. Landw. Gesell., Bd. 16, No. 10, p. 55–56; No. 11, p. 57–60.

(250) WAGNER, P., AND DORSCH 1892 Die Stickstoff Dungung der Landwirschaftlichen Kulturpflanzen. Berlin.

(251) WAGNER, P., et al 1907 Versuche über die Stickstoff dungung der Kulturpflanzen. *In* Arb. Deut. Landw. Gesell., No. 129, p. 6–286.

(252) WAGNER, P., et al 1908 Fertilizer experiments with tobacco. *In* Arb. Deut. Landw. Gesell., No. 138, p. 99.

(253) WARD, R., et al 1907 Agricultural experiments. *In* Rpt. Bot. Gard. and Onderneem School Farm Brit. Guiana, 1906–7, p. 1–13.

(254) WARINGTON, R. 1900 The comparative value of nitrate of soda and sulfate of ammonia as manures. *In* Jour. Roy. Agr. Soc. England, ser. 3, v. 11, pt. 2, no. 42, p. 300–346.

(255) WATERS, H. J., AND HESS, E. H. 1894 General fertilizer experiments. *In* Penn. Agr. Exp. Sta. Ann. Rpt. 1894, pt. 2, p. 258–281

(256 WATTS, F., et al 1908 Manurial experiments. *In* Imp. Dept. Agr. West Indies. Sugar-Cane Expts. Leeward Isl., pt. 2, p. 22.

(257) WATTS, F., et al 1911 Experiments with field crops. *In* Rpts. Bot. Sta. (etc.) St. Kitts-Nevis, 1910–11, p. 9–13, 15–33.

(258) WEHNERT, 1913 Comparative fertilizer experiments with cereals in 1912. *In* Landw. Wchnbl. Schles.-Holst., Bd. 63, No. 51, p. 1010–1016.

(259) WEIBULL, M., et al 1907 Cooperative fertilizer trials in Malmohus County, 1906. *In* Malmö. Läns K. Hushall. Sällsk. Kvrtlsskr., no. 1, p. 197–265.

(260) WEIN, E. 1905 The nitrogen feeding of agricultural plants. *In* Chem. Ztg., Bd. 29, No. 80, p. 1066, 1067.

(261) WHEELER, H. J., AND ADAMS, G. E. 1908 A further study of soil treatment in greenhouse culture. *In* R. I. Agr. Exp. Sta. Bul. 128, p. 183–194.

(262) WHITE, J. W. 1913 The results of long-continued use of ammonium sulfate upon a residual limestone soil of the Hagerstown series. *In* Penn. Agr. Exp. Sta. Ann. Rpt. 1913, p. 55–104.

(263) WIDTSOE, J. A. 1912 The production of dry matter with different quantities of irrigation water. Utah Agr. Exp. Sta Bul. 116, p. 1–64.

(264) WIDTSOE, J. A., AND MERRIL, L. A. 1912 The yields of crops with different quantities of irrigation water. Utah Agr. Exp. Sta. Bul. 117, p. 65–119.

(265) WILCOX, E. V., KELLEY, W. P., AND KRAUSS, E. G. 1910 Work with field crops at the Hawaii Station. *In* Hawaii Agr. Exp. Sta. Ann. Rpt. 1910, p. 12–14; 43, 44, 51–64.

(266) WOLKOFF, M. I. 1917 Adsorption of ammonium sulfate of different concentrations by different soils and quartz sand of different fineness. *In* Soil Sci., v. 3, no. 6, p. 561–564.

(267) WOLKOFF, M. I. 1918 Effect of ammonium sulfate in nutrient solution on the growth of soybeans in sand cultures. *In* Soil Sci., v. 5, no. 2, p. 123–150.

(268) WOLLNY, E. 1897 Untersuchung den Einfluss der Wachsthumsfaktoren auf des Produktionsvergogen der Kulturpflanzen. *In* Forsch. Geb. Agr. Phys., Bd. 20, p. 53–109.

(269) WOODS, C. D. 1915 Field experiments. Me. Agr. Exp. Sta. Bul. 236, p. 41–56, 63, 64.

(270) WRIGHT, R. P. 1911 Report on experiments on the manuring of potatoes in 1907 and 1908. *In* West of Scot. Agr. Col. Ann. Rpt. 10, p. 107–132.

ON THE EXTRACTION OF AMMONIA FROM SOIL

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INTRODUCTION

During the course of experiments conducted at the Ohio Agricultural Experiment Station upon the relationship existing between ammonification and other processes in soils, a comparison of the amounts of ammonia extracted from soils by water and 5 per cent hydrochloric acid was made. The results obtained are briefly outlined in this paper.

In a series of soil mixtures including treatments having an influence upon the amount of ammonia gradually being produced by the action of ammonifying organisms, upon added materials such as casein, the fixation of ammonia might be greater than if the total amount of ammonia formed was present at any one time. A water extract of these soils will not give the full amount of ammonia in them, but if it gives a proportional amount to that found by either distillation with magnesium oxide or extraction with acid, then for comparative purposes the ammonia found in the water extract would be just as useful as the larger amounts found by distillation or acid extraction.

McBeth (2) has recently shown that by making an acid extraction of soils he could obtain all the ammonia that could be obtained by distilling the soil with magnesium oxide, and at the same time get better-checking duplicates than could be obtained by distilling with MgO.

EXPERIMENTAL

The soils used were all surface soils (0–7 inches) which had been air-dried and ground sufficiently fine to pass through a 2-mm. sieve. Three distinct types of soil were included in this work; an acid silt loam, which is deficient in bases and organic matter, a basic black clay, well supplied with organic matter, and a very acid peat soil. These are designated as Wooster silt loam, Paulding clay, and peat.

While the data reported were chiefly obtained on different soil mixtures used in the investigations previously referred to, these same soils were also used in a preliminary experiment to determine the percentage of added ammoniacal nitrogen furnished by ammonium sulfate which could be recovered by extraction with water and with acid.

Methods

Portions of the soils were placed in 1-liter bottles and ammonium sulfate sufficient to supply 0.1484 gm. of nitrogen was added to half of the bottles. Water or 5 per cent hydrochloric acid was added at the rate of 500 cc. per 100 gm. of soil and extraction continued for 30 minutes with constant shaking in a machine. Nitrogen as ammonia was determined by distilling duplicate 200-cc. portions with 0.5 gm. of freshly-calcined magnesium oxide, first exactly neutralizing the acid extracts with sodium hydroxide.

The distillation was made in 500-cc. flasks, the distillate first passing through a second and smaller flask used as a scrubber, and from this through Pyrex glass tubing into Pyrex Erlenmeyer flasks; Pyrex glass being found as satisfactory as quartz for this purpose. As the acid in the receiving flask became very hot, the flasks were cooled under a water tap before the back titration was made with N/20 sodium hydroxid solution, methyl red being used as the indicator.

TABLE 1

Recovery of added nitrogen by water and 5 per cent hydrochloric acid

SOIL	NITROGEN RECOVERED							
	By Water				By Acid			
	Nitrogen from soil alone	From soil +0.1484 gm. nitrogen in ammonium sulfate	Increase	Per cent recovered	Nitrogen from soil alone	From soil +0.1484 gm. nitrogen in ammonium sulfate	Increase	Per cent recovered
	gm.	*gm.*	*gm.*		*gm.*	*gm.*	*gm.*	
Wooster silt loam.....	0.0	0.0941	0.0941	63.41	0.0073	0.1562	0.1489	100.00
Paulding clay.....	0.0	0.0586	0.0586	39.48	0.0117	0.1437	0.1320	88.94
Peat.....	0.0073	0.0949	0.0876	59.48	0.0155	0.1355	0.1200	80.86

The results obtained by the above treatments are given in table 1.

The water extraction has failed to give more than 60 per cent of the added ammonia, while the acid extraction gave complete recovery in one case and more than 80 per cent in the other two cases.

Further and more conclusive data were obtained by determining both water and acid-soluble ammonia produced in a series of differently-treated soils that were being used in an investigation pertaining to the relation between sulfofication, nitrification, and ammonification in soils (1).

The different treatments for the Wooster soil with the water and acid-soluble ammonia obtained are given in table 2.

In this series of soils, twenty-seven 500-gm. portions of Wooster soil were weighed out into quart jars on March 16, 1917, and the treatments added as designated in the tabulation of results. These soils were kept under control so that optimum conditions of temperature and moisture prevailed. After

standing for a period of 17 weeks, the soils were thoroughly mixed and 400 gm. of each was extracted for 16 hours with 2500 cc. of distilled water free from ammonia and nitrates. These soil solutions were filtered through Berkfield filters, with an air pressure of 20 pounds. The solutions obtained were clear and free from sediment.

TABLE 2

Nitrogen as parts per million of soil, extracted from differently-treated samples of Wooster silt loam

TREATMENT			AMMONIACAL NITROGEN		INCREASE OF ACID OVER WATER EXTRACTION
Sulfur	Casein	Calcium carbonate	Extracted with water	Extracted with acid	
gm.	*gm.*	*gm.*	*p. p. m.*	*p. p. m.*	*p. p. m.*
0.0	0.00	0.0	0.0	0.0	0.0
0.0	1.75	0.0	132.3	196.0	64.0
0.0	0.00	2.0	0.0	0.0	0.0
0.0	1.75	2.0	0.0	14.0	14.0
0.0	0.00	0.0	0.0	0.0	0.0
0.0	1.75	0.0	131.2	183.0	52.0
0.0	0.00	2.0	0.0	0.0	0.0
0.0	1.75	2.0	0.0	10.5	10.5
0.5	0.00	0.0	32.8	60.0	32.2
0.5	1.75	0.0	328.0	416.0	88.0
0.5	0.00	2.0	0.0	0.0	0.0
0.5	1.75	2.0	0.0	51.0	51.0
0.5	0.00	1.0	28.4	45.5	17.1
0.5	1.75	1.0	240.6	324.0	84.0
0.5	0.00	0.5	45.0	47.0	2.0
0.5	1.75	0.5	207.7	306.0	99.0
0.5	0.00	0.0	48.1	89.0	41.0
0.5	1.75	0.0	328.1	455.0	127.0
0.5	0.00	2.0	0.0	0.0	0.0
0.5	1.75	2.0	87.5	131.0	43.5
0.5	0.00	1.0	37.1	70.0	32.9
0.5	1.75	1.0	278.0	378.0	100.0
0.5	0.00	0.5	42.5	87.0	44.5
0.5	1.75	0.5	288.7	390.0	103.0

All results are averages of duplicate determinations. A comparison of the amounts of ammonia extracted by water and acid from the soil with and without the addition of casein is shown in table 3.

When casein was not included in the treatment added to the soil, the amount of nitrogen as ammonia found by both the water and acid methods of extraction is much less than when casein was included in the treatment. With the smaller amounts of ammonia found in the soil without casein the acid extraction shows a high percentage increase, the acid extracting about 67 per cent more nitrogen as ammonia than did water. That this increase is not due to the acid extracting nitrogen from other than ammoniacal compounds is evident

from the fact that in six of the twelve samples, no nitrogen as ammonia was extracted by either water or acid treatment. In these six samples conditions were favorable for nitrification and any ammonia produced was changed to nitrates.

TABLE 3

Comparison of ammonia extracted from Wooster silt loam with and without the addition of casein by water and 5 per cent hydrochloric acid

TREATMENT			WATER—EXTRACTED AMMONIA	ACID—EXTRACTED AMMONIA	INCREASE OF ACID OVER WATER EXTRACTION	PER CENT INCREASE
Sulfur	Casein	Calcium carbonate				
With casein						
gm.	*gm.*	*gm.*	*p. p. m.*	*p. p. m.*	*p. p. m.*	
0.0	1.75	0.0	132	196	64	48.4
0.0	1.75	2.0	0	14	14	14.0
0.0	1.75	0.0	131	183	52	39.6
0.5	1.75	2.0	0	10	10	10.0
0.5	1.75	0.0	328	416	88	26.8
0.5	1.75	2.0	0	51	51	51.0
0.5	1.75	1.5	240	324	84	35.0
0.5	1.75	0.0	207	306	99	47.8
0.5	1.75	0.0	328	455	127	38.7
0.5	1.75	2.0	87	131	43	49.7
0.5	1.75	1.0	278	378	100	35.9
0.5	1.75	0.5	288	390	103	35.7
Without casein						
0.0	0.0	0.0	0	0	0	0.0
0.0	0.0	2.0	0	0	0	0.0
0.0	0.0	0.0	0	0	0	0.0
0.5	0.0	2.0	0	0	0	0.0
0.5	0.0	0.0	32	59	27	83.0
0.5	0.0	2.0	0	0	0	0.0
0.5	0.0	1.0	24	45	21	87.0
0.5	0.0	0.5	45	47	2	4.4
0.5	0.0	0.0	48	89	41	85.2
0.5	0.0	2.0	0	0	0	0.0
0.5	0.0	1.0	37	70	32	88.6
0.5	0.0	0.5	44	87	43	104.7

In the mixtures where ammonia was found, the oxidation of sulfur included in the treatment has depressed nitrification and the change from proteid to nitric nitrogen did not proceed completely, part of the ammonia formed remaining as ammonium sulfate. In each of these samples the ratio of the water-extracted ammonia to that extracted with acid is the same in each case, being 1.8 parts by acid for every part by water.

When casein was included in the treatments added to the soil, the amount of nitrogen as ammonia found was greatly increased and the amount extracted by water approaches that extracted by acid.

As in the case of the soils without added casein when conditions were favorable for nitrification, the amount of ammonia found is small, but when the acidity developed by the oxidation of added sulfur has not been neutralized by calcium carbonate, the amount of ammonia found is high. In eight such cases the ratio of the water to acid-extracted ammonia is 1.3 parts by acid extraction to each part extracted with water.

Extraction of ammonia from Paulding clay

This soil is a very basic clay soil and differs very much from the Wooster silt loam. No casein was added to this soil, as it contains much natural organic matter. The results with this soil are found in table 4.

TABLE 4

Extraction of ammoniacal nitrogen from basic Paulding clay

TREATMENT		AMMONIACAL NITROGEN		INCREASE OF ACID OVER WATER EXTRACTION
Sulfur	Calcium carbonate	Extracted with water	Extracted with acid	
gm.	*gm.*	*p. p. m.*	*p. p. m.*	*p. p. m.*
0.0	0.0	10.9	78	67.1
0.0	0.5	10.9	56	45.1
0.0	0.0	10.9	35	24.1
0.0	0.5	0.0	52	52.0
0.5	0.0	10.9	44	33.1
0.5	0.5	10.9	47	36.1
0.5	0.25	9.8	64	44.2
0.5	0.0	9.8	78	68.2
0.5	0.5	10.9	52	41.1
0.5	0.25	16.9	54	37.1

With this basic clay soil which contains 10 parts per million of water-soluble nitrogen as ammonia, the 5 per cent hydrochloric acid solution has been able to extract an average of 50 parts per million of nitrogen.

With this clay soil the ratio of the water- to the acid-extracted ammonia is 4.6 parts of acid-extracted ammonia for every part extracted by water.

Extraction from peat

The peat used was an acid peat of rather raw texture. The solution obtained from this soil was highly colored but without sediment. The results for the peat are given in table 5.

The high figures obtained for ammonia from this peat by acid extraction might cause one to believe that other than ammonium compounds had been extracted by the acid, but it must be remembered that in the preliminary work, the 5 per cent hydrochloric acid was able to recover only 80 per cent of the added nitrogen from this peat.

TABLE 5

Extraction of ammoniacal nitrogen from an acid peat

TREATMENT		AMMONIACAL NITROGEN		INCREASE OF ACID OVER WATER EXTRACTION	PER CENT INCREASE
Sulfur	Calcium carbonate	Extracted with water	Extracted with acid		
gm.	*gm.*	*p. p. m.*	*p. p. m.*	*p. p. m.*	
0.00	0.00	259	525	266	102.7
0.00	1.50	122	210	88	72.1
0.00	0.00	252	532	280	111.1
0.00	1.50	115	203	88	76.5
0.15	0.00	357	598	241	67.5
0.15	1.50	399	684	285	71.4
0.15	0.75	406	595	189	46.5
0.15	0.30	402	595	193	48.0
0.15	0.00	392	630	238	60.7
0.15	1.50	318	630	312	98.1
0.15	0.75	420	700	280	66.6
0.15	0.30	403	626	223	55.3
0.00	0.00	252	544	292	115.8

SUMMARY

While in no case has the water extraction given the full amount of ammonia from the different soils, yet in each group a certain ratio appears to exist between the amounts extracted by the two methods, and for comparative use, the ammonia found in the water extract would be just as useful as the somewhat larger amount found by extracting the soil with 5 per cent hydrochloric acid.

REFERENCES

(1) Ames, J. W., and Richmond, T. E. 1918 Sulfofication in relation to nitrogen transformations. *In* Soil Sci., v. 5, no. 4, p. 311–321.

(2) McBeth, I. G. 1917 Fixation of ammonia in soils. *In* Jour. Agr. Res., v. 9, no. 5, p. 141–155.

THE OCCURRENCE OF BACTERIUM LACTIS VISCOSUM IN SOIL

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Little is known concerning the natural habitat of most microörganisms, hence any contribution to this phase of bacteriology will furnish welcome and valuable information to students of this subject.

During the autumn of 1915 at the New Jersey Agricultural Experiment Station, in the course of certain bacteriological investigations of soil, a colony resembling *Bacterium lactis viscosum* was found on a plate of beef broth agar. The plate had been poured from a dilution of 1 to 50,000. The soil was a Sassafras sandy loam which had not been manured with cow dung for at least 3 years. The writer's attention was directed to this colony on account of its great viscosity and its reddish tint. After isolation in pure culture on agar slants, its morphological and cultural characteristics were determined, including its action on milk. The organism corresponded with Conn's description in every particular except that in the soil culture the reddish color was somewhat more pronounced on solid media. It also agreed very well with the stock laboratory culture of *Bact. lactis viscosum*.

The next year (1916), another organism of this type was isolated from a large decayed soybean nodule. It made appreciable growth on Ashby's mannite agar, which is nearly nitrogen-free. On account of the very viscid nature of the colony it was transferred to broth agar and the cultural characteristics of the bacterium determined in the usual way. These tests confirmed the indication that the organism was *Bact. lactis visocsum*. It produced some pigment but this character was not nearly as pronounced as in the organism first isolated. After three transfers on broth agar, it was replanted on Ashby's mannite agar but no growth was observed. The organism had apparently lost its power of living on this nitrogen-poor medium. Perhaps like many other freshly-isolated soil organisms it is able for a short time to fix some atmospheric nitrogen.

The soybean field from which this second organism was isolated, was located within a hundred yards of a heap of horse manure but no cow dung had been added to the soil in several years. Cattle had not been allowed to roam on either field for over twenty years. The soil on which the soybeans grew was a Penn shaley loam.

The presence of this organism in soils of two different types, indicates that the soil is probably a natural medium for the development of this widely

distributed and trouble-causing organism. From at least three cases of slimy milk in the state of New Jersey during the past two years, identical cultures of this organism were obtained. If the soil is a home of this bacterium, then the sporadic outbreaks of ropy milk in dairies throughout the country may be more readily understood. At first usually but one teat of the cow is affected, then it spreads rapidly to the other teats or other animals unless promptly checked. Unless all utensils which come in contact with ropy milk are thoroughly sterilized, they may harbor the organisms and be a potent source of trouble for a long time. The organism does not form spores, but there are many other non-spore-forming bacteria in the soil. Perhaps the slime produced serves as a sort of protection and aids it to multiply and live in the soil.

SUMMARY

Two organisms, corresponding to written descriptions and laboratory cultures of *Bact. lactis viscosum*, were isolated from two different soils, which had not received applications of cow dung for several years. It is probable that the soil is a natural habitat of this organism.

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